# Integrating anthropometric and cardiometabolic health methods in stress, early experiences, and development (SEED) science

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#### Abstract

Within Stress, Early Experiences, and Development (SEED) science, there is a growing body of research demonstrating complex associations not only between stress, development, and psychopathology, but also with chronic disease risk factors. We argue that it is important for SEED researchers to consider including child anthropometric and physical health measures to more comprehensively capture processes of risk and resilience. Broader adoption of harmonized anthropometry and health measures in SEED research will facilitate collaborations, yielding larger datasets for research in high-risk populations, and greater opportunity to replicate existing findings. In this review, we identify optimal anthropometric and cardiometabolic health measurement methods used from infancy through adolescence, including those that are low-burden and inexpensive. Methods covered include: waist, hip, and head circumference, height, length, weight, pubertal development, body composition, blood pressure, arterial stiffness, carotid intima media thickness, and serum measures of cardiometabolic risk and inflammation. We provide resources for SEED researchers to integrate these methods into projects or to better understand these methods when reading the literature as well as where to find collaborators for more in-depth studies incorporating these measures. With broader integration of psychological and physical health measures in SEED research, we can better inform theory and interventions to promote health and resilience in individuals who have experienced early stress.

#### KEYWORDS

anthropometry, body composition, cardiometabolic health, development, inflammation, puberty, stress

### 1 | INTRODUCTION

Research in the area of stress, early experiences, and development (SEED) is becoming increasingly interdisciplinary, integrating psychology, pediatrics, public health, biology, sociology, technology, and other disciplines. This trend is particularly exciting, as it broadens the research questions and types of problems researchers will be able to solve. For example, cardiovascular disease (CVD) is the number one cause of death for men and women (Heron, 2018), and child obesity rates in both developed and developing countries are alarmingly high (World Health Organization, 2018). Meanwhile, global malnutrition remains a problem, with around 155 million children experiencing growth stunting, which has major implications for brain development and health (World Health Organization, 2018). Yet, these fields have traditionally been the focus of medical and public health researchers and not psychologists and sociologists. Yet, behavioral and psychosocial risk factors are key determinants of cardiovascular risk (Cappuccio, Cooper, D'Elia, Strazzullo, & Miller, 2011; Halonen et al., 2015; Low, Matthews, Kuller, & Edmundowicz, 2011; McDade, Hawkley, & Cacioppo, 2006; Warren et al., 2010). Now, social scientists are more likely to pursue questions about how psychosocial stress and other socioemotional and behavioral factors contribute to CVD. In addition, transdisciplinary work on cardiovascular risk has benefited through theories from or informed by the social sciences, such as the Developmental Origins of Health and Disease theory (Gluckman & Hanson, 2006), the allostatic load model (McEwen, 1998), and biological sensitivity to context theory (Boyce & Ellis, 2005), among others. In addition, methodological tools such as careful measurement of psychosocial stress (Stress Measurement Network, 2020), mental health constructs (Van der Kooy et al., 2007), and cognitive (O'Donovan, Slavich, Epel, & Neylan, 2013) and emotional processes (Lovallo & Gerin, 2003) have been highly important for better understanding the psychosocial precursors of CVD. Increasingly transdisciplinary research means that tools used in other fields can be used to examine questions usually in the domain of social scientists. For example, anthropometrics (measurement of the body) and measures of cardiovascular health are being integrated into SEED research on normal and abnormal development.

# 2 | GOALS AND OUTLINE OF THIS REVIEW

The goal of the current review is to provide a methodological overview and a number of resources for implementing measures of anthropometry and cardiometabolic health into research. We encourage social scientists to integrate these methods into SEED science and form transdisciplinary collaborations. We also want this review to serve as a guide for SEED researchers who want to better understand these measures when they read papers incorporating anthropometry, nutrition status, and cardiometabolic health. First, we discuss the Developmental Origins of Health and Disease theory, which guides much of the research on prenatal and postnatal factors influencing long-term health. Second, we briefly review interdisciplinary literature that connects social science and anthropometry, nutrition status, and cardiometabolic health to show the value of adding these methods to SEED research (see Table 1). We next review the following methods used from infancy through childhood and adolescence: anthropometry, more advanced techniques for measuring body composition, and ways to measure cardiovascular health. Anthropometry includes measuring height/length, weight, and waist, hip and head circumference. Body composition may be assessed by measurement of skin fold thickness, bioelectrical impedance, air displacement plethysmography, and dual-energy X-ray absorptiometry. Measurements related to cardiovascular health include blood pressure, arterial stiffness, carotid intima media

 
 TABLE 1
 Reasons SEED scientists should assess anthropometry and cardiometabolic health

- Why should SEED scientists assess anthropometry and cardiometabolic health?
- 1. There are bidirectional associations between anthropometry, physical health, and developmental outcomes (Chrousos, 2009; Grantham-McGregor et al., 2007; Liang et al., 2014; Lozoff et al., 2006; Oriá et al., 2016; Prince et al., 2007; Walker et al., 2007)
- 2. Deficiencies or acceleration in physical growth and health problems may be an indicator of a maladaptive environment that could negatively affect brain development, such as malnutrition or high psychosocial stress (Doom & Georgieff, 2016; Grantham-McGregor et al., 2007; Lozoff et al., 2006; Walker et al., 2007)
- 3. Including anthropometry and measures of cardiometabolic health in studies will increase our progress in finding mediators and moderators of associations between anthropometry, health, and developmental outcomes. Even if the main, moderating, or mediating effects of early stress on anthropometry and cardiometabolic health are not the primary focus of investigators, these factors are important control variables to make sure investigators are isolating their constructs of interest
- 4. With the push for more open and reproducible science, data sharing will allow SEED researchers to compile large datasets so that those interested in anthropometry and health can use these data for sufficiently-powered studies (Nosek et al., 2015). Especially when working with hard-to-access populations, which is often the case in SEED science, data can be shared with others who are trying to understand health and physical growth in these populations
- 5. Psychological science has become more interdisciplinary (Michel, Marcinowski, Babik, Campbell, & Nelson, 2015). We are learning more about associations between anthropometry, health, and developmental outcomes, which increase opportunities to translate psychological concepts to medicine and public health, among other fields, to improve lives. We can incorporate these methods into psychological studies to inform our theories of development and to seek funding for research with implications outside the social sciences. Interdisciplinary research increases opportunities to create, test, and disseminate effective interventions (Cicchetti & Blender, 2006)

thickness, and serum measures including lipids, glucose, insulin, and C-reactive protein. We review gold-standard methods and compare them to low-cost, quick and minimally invasive methods (see Table 2).

For investigators who would like to focus their SEED research on anthropometry, nutrition status and cardiometabolic health, we review more in-depth methods of assessment. We provide a decision tree in Figure 1 to help researchers decide which methods might be best for their studies, as there are trade-offs that must be considered with respect to time, funding, participant burden, and access to facilities with advanced cardiometabolic assessment methods. We have included methodological suggestions, sources of in-depth information on specific methods and suggestions for finding methods experts to collaborate with. While this is not a comprehensive overview of all possible methods that could be used, the selected measures are directly applicable to SEED scientists. For example,

#### TABLE 2 Methods for assessing anthropometry and cardiometabolic health from infancy to adolescence

				Ideal for age group		
	Ease of implementation	Cost	Participant burden	Infants (age 0–24 months)	Children (age 2–10 years)	Adolescents (age 11–18 years)
Anthropometry						
Height	Easy	\$	Low		Х	Х
Length	Easy	\$	Low	Х		
Weight	Easy	\$	Low	Х	Х	Х
Waist circumference	Easy	\$	Low		Х	Х
Head circumference	Easy	\$	Low	Х	Х	Х
Self-report of pubertal status	Easy	\$	Low		Х	Х
Trained assessor's examination of pubertal status	Difficult	\$\$\$\$	Medium		Х	Х
Body composition						
Skinfold measurement	Medium	\$\$	Medium	Х	Х	Х
Bioelectrical impedance analysis (BIA)	Easy	\$\$\$	Low		Х	Х
Air displacement plethysmography (ADP)	Medium	\$\$\$\$	Medium	Х	Х	Х
Dual-energy X-ray absorptiometry (DXA)	Medium	\$\$\$\$	Medium	х	х	Х
Cardiometabolic risk						
Blood pressure	Medium	\$\$	Medium		х	х
Serum measures of cardiometabolic risk and inflammation	Medium-to- Difficult	\$\$-\$\$\$	High		Х	Х
Arterial stiffness	Medium	\$\$\$\$	Medium		Х	Х
Carotid intima media thickness	Difficult	\$\$\$\$	Medium			Х

*Note*: Ease of implementation includes the need to train and/or hire individuals to obtain the measure (e.g., phlebotomist for serum samples, assessor for accurate and reliable puberty assessment). Cost ranges from less than 200 (, 200-1,000 (, 1,000-5,000 (, 1,000

although relevant to growth and cardiometabolic health, we do not review autonomic nervous system (ANS) or hypothalamic-pituitaryadrenal (HPA) measures in the current paper as there are excellent reviews of ANS (van Dijk et al., 2013; Mendes, 2009) and HPA methodology (Adam & Kumari, 2009; Jessop & Turner-Cobb, 2008) in the literature. For the purpose of this review, we refer to infancy as the period before 24 months of age, childhood from 2 to 9 years, and adolescence from 10 to 19 years.

### 3 | DEVELOPMENTAL ORIGINS OF HEALTH AND DISEASE THEORY

Central to developmental science is the importance of theory for guiding research. A leading theory guiding study of the early life predictors of cardiovascular health problems is the Developmental Origins of Health and Disease (DOHaD), also known as the Barker hypothesis. David Barker's epidemiological work beginning in the 1980s demonstrated that fetal undernutrition is a robust predictor of later life coronary heart disease (Barker, 1995; Barker & Osmond, 1986). Undernutrition during fetal life is associated with biological programming of a "thrifty phenotype" that leads to higher weight and greater likelihood of cardiometabolic diseases, particularly in a postnatal environment with greater access to high-calorie foods (Vaag, Grunnet, Arora, & Brøns, 2012). Research on DOHaD has expanded from a focus on just fetal nutrition and adult CVD to focus on both prenatal and postnatal factors, including environmental exposures, stress exposure, and relationships, that affect a wide range of adult diseases (Suzuki, 2018). The DOHaD framework is particularly important for developmental scientists as it is known that socioeconomic and psychosocial factors such as food insecurity and maternal mental health are associated with fetal development and nutrition (Field, Diego, & Hernandez-Reif, 2006; Laraia, Siega-Riz, & Gundersen, 2010). In addition, postnatal factors from infancy through adolescence, such as stressful life experiences, social relationships, and nutrition are also associated with adult health (Cohen,



**FIGURE 1** Decision tree for planning which growth and cardiometabolic risk measures may be the best for a study. %BF, percent body fat; ADP, air displacement plethysmography; BIA, bioelectrical impedance analysis; CM risk, cardiometabolic risk; DBS, dried blood spot; DXA, dual-energy X-ray absorptiometry

Janicki-Deverts, Chen, & Matthews, 2010; Puig, Englund, Simpson, & Collins, 2013; Victora et al., 2008). As the application of DOHaD theory has expanded to additional exposures, outcomes, and developmental periods, the hypothesized mechanisms that have been studied using this theory have also expanded. For example, early studies focused on insulin resistance, lipid metabolism, and blood pressure alterations as potential primary mechanistic pathways of interest between fetal undernutrition and adult cardiometabolic disease (Barker, 1995). More recent mechanisms of interest between pre- and postnatal factors and adult health include alterations in epigenetics, the ANS, the HPA axis, the immune system, the metabolic system, brain development, and behavior, among many others (e.g., Danese & Lewis, 2017: Matthews & McGowan, 2019: Wadhwa, Buss, Entringer, & Swanson, 2009). DOHaD has served as the guiding theory for much of the work in the area of developmental programming of cardiometabolic risk, and as a result, it serves as the guiding theory for this methodological review.

### 4 | ANTHROPOMETRY, NUTRITION STATUS, CARDIOMETABOLIC HEALTH, AND SOCIAL SCIENCE

# 4.1 | Anthropometry, nutrition status, and SEED science

Within the DOHaD framework, many prenatal and postnatal factors contribute to growth and cardiometabolic risk across the lifespan. For example, body size and shape are determined by a number of factors, including genetics, the microbiome, the intrauterine environment, food access, the built environment, diet, physical activity, and social factors. Any of these factors may impart risk, which may present as deficient or excess physical growth. For example, nutrients are allocated for different purposes, including physical growth, brain development, and providing energy for bodily processes. Nutrients include macronutrients (e.g., fats, carbohydrates, and protein), which are components of daily calorie intake and more closely associated with physical growth, and micronutrients (e.g., iron, Vitamin D, zinc, and choline). Different types and amounts of nutrients are needed at different developmental stages. Inadequate nutrient intake, especially too few calories, commonly leads to stunting (suboptimal growth in height/length) when more chronic, and wasting (suboptimal growth in weight-for-length or weight-forheight) when more acute, and can also lead to small head circumference (microcephaly) (Gahagan, 2006; Gahagan & Holmes, 1998). Undernutrition also leads to problems with brain development since the brain is metabolically costly in early life during rapid neurodevelopment. Psychosocial stress, through the activation of stress-mediating systems, can interfere with nutrient absorption and nutrient trafficking within the body, even with adequate intake of nutrients (Monk, Georgieff, & Osterholm, 2013; Suchdev et al., 2017).

Physical growth can be a useful metric of development in part because it is metabolically demanding. Disruptions in physical growth trajectories can affect nutrients available for brain development, leading to disruptions in cognitive, social, and emotional development that can last into adulthood despite treatment (Lozoff et al., 2006). Psychosocial stress inhibits physical growth, partly mediated through HPA axis function (Chrousos, 2009). Children experiencing chronic psychosocial stress often show impairments in bone growth, including height/length and head circumference, which rebound when conditions improve (Johnson, Guthrie, et al., 2010). This growth failure in children experiencing chronic psychosocial stress is typically mediated by chronic inadequate caloric intake, which may be due to insufficient food for the child or their inability to consume or absorb sufficient calories. Thus, SEED scientists interested in psychosocial stress across the lifespan should consider the combined effects of psychosocial stress and nutritional deficiencies on neurodevelopment and developmental outcomes. Anthropometry can provide additional information about environmental deprivation in the household, as it is relatively sensitive to acute and chronic nutritional deficits. Psychosocial stress may also accelerate growth processes such as pubertal development, leading to faster gains in weight and linear growth. As a result, there are U-shaped associations between psychosocial stress and growth across development where increased stress can lead to overweight or underweight, or to increased or decreased linear growth.

#### 4.1.1 | Height/length/weight deficits

Prenatal physical growth restriction has demonstrated associations with child neurodevelopment (Levine et al., 2015) and is generally measured using birth weight adjusted for gestational age at birth. Infants born small for gestational age (SGA) may be constitutionally small (e.g., due to the effects of genes) or may have experienced intrauterine growth restriction (IUGR), which is a pathological restriction of genetic growth potential in utero. SGA infants are those with low birth weight for gestational age regardless of their rate of intrauterine growth, while IUGR describes the rate of intrauterine growth regardless of eventual birth weight and is considered a pathological process. IUGR is present in approximately 10%-15% of all pregnancies, with variation by the population assessed (Longo et al., 2013). IUGR can be due to maternal, placental, fetal, nutritional, or other environmental factors that slow fetal growth (Sharma, Shastri, & Sharma, 2016). Infants who are SGA show poorer cognitive functioning and are more likely to have atypical neurologic status compared to infants who are appropriate for gestational age (AGA) (McCarton, Wallace, Divon, & Vaughan, 1996). IUGR may cause these developmental outcomes, or it could be that problems leading to IUGR such as maternal or fetal factors could lead to both poor developmental outcomes and IUGR. SGA and IUGR infants are more likely than their normal birth weight and unrestricted counterparts to show lower developmental quotients, delays in problem-solving, and delayed motor development (Doom & Georgieff, 2016; Grantham-McGregor, Lira, Ashworth, Morris, & Assunçao, 1998; Latal-Hajnal, von Siebenthal, Kovari, Bucher, & Largo, 2003). These effects can last into adolescence and adulthood.

Linear growth stunting, defined as height- or length-for-age two standard deviations (SDs) below the mean on international reference growth curves, is used as an indicator of early developmental risk (Grantham-McGregor et al., 2007). Stunting and developmental problems may have common causes. A longer duration of stunting during childhood may be more harmful for child development than stunting for a short duration (Doom & Georgieff, 2016). Earlier stunting may also have more harmful effects than later stunting, as infant brain development is rapid and tends to track growth in length. Thus, the timing and duration of stunting is important to consider, particularly in high-risk populations. Even after controlling for socioeconomic status, stunting is associated with delays in cognitive development, poorer school achievement, and greater risk for failing to enroll in or dropping out of school (Doom & Georgieff, 2016; Walker, Chang, Powell, & Grantham-McGregor, 2005). One analysis estimates that early stunting is associated with a 22.2% reduction in adult income for those who were stunted but not living in poverty in childhood and a 30.1% reduction for those who were both stunted and living in poverty (Grantham-McGregor et al., 2007). Both stunting and low weight-for-age are associated with less positive effect, less play, greater lethargy, and greater risk of insecure attachment compared to non-growth delayed young children (Gardner, Walker, Powell, & Grantham-McGregor, 2003; Graves, 1978). Adolescents who were stunted in childhood also show more symptoms of depression, anxiety, hyperactivity, and lower self-esteem compared to adolescents who were not stunted (Walker, Chang, Powell, Simonoff, & Grantham-McGregor, 2007). Consistent with the DOHAD model, there is also evidence that growth stunting early in life is associated with higher blood pressure (Gaskin, Walker, Forrester, & Grantham-McGregor, 2000), abdominal fatness (Schroeder, Martorell, & Flores, 1999), and cardiometabolic risk (DeBoer et al., 2012).

#### 4.1.2 | Head circumference

Smaller head circumference can be associated with malnutrition and suboptimal brain development, especially during childhood. However, head circumference is not always included as an indicator of dietary intake on brain development in studies of high-risk children. Previous research has demonstrated associations between head circumference, rate of head growth, the environment, and developmental outcomes. Within a cohort of very low birth weight children (<1.5 kg at birth), those children with head circumference more than 2 *SD* below the reference population mean at 8 months showed greater neurologic impairment, and lower scores for receptive language, speech, verbal and performance intelligence quotient (IQ), academic achievement in reading, spelling, and math, and greater hyperactivity at 8 years compared to very low birth weight children with normal head circumference (Hack et al., 1991). Head growth over time may be an indicator of psychosocial or nutritional risk that can have underlying effects on child development. Impaired head growth and developmental problems may also have common causes.

#### 4.1.3 | High-risk populations and catch-up growth

High-risk populations include those experiencing nutritional deprivation, socioeconomic hardship, and/or high levels of psychosocial stress. Anthropometry is particularly important to measure in highrisk populations as nutritional deficiencies and psychosocial stress, both of which are associated with restricted growth, are more prevalent in these populations (though both stunting and wasting also occur in low-risk, highly-resourced families). Children in specific contexts of stress, such as institutional care and foster care, often show growth delays in height/length, weight, and head circumference. Most of these children display remarkable catch-up growth in height/length and weight, but less so in head circumference, when their environments vastly improve (Van IJzendoorn, Bakermans-Kranenburg, & Juffer, 2007). Because of nutritional deficiencies and psychosocial risk for children in contexts such as institutional care or foster care, care providers must be aware that these needs must be addressed and that rapid catch-up growth may put children at risk for greater nutritional deficiencies such as iron deficiency if the specific nutrient intake is not sufficient to support the accelerated growth rate (Tooley, Makhoul, & Fisher, 2016).

Catch-up growth is an important measure for both researchers and practitioners, as the degree of catch-up growth is associated with a number of developmental outcomes. Catch-up growth following growth restriction in infants may be more closely associated with neurodevelopment than size for age (Doom & Georgieff, 2016). A meta-analysis of data from low- and middle-income countries demonstrates robust associations between height/length and cognitive ability, with each additional unit of length-for-age z-score associated with a 0.24 SDs higher score for children ≤2 years old, and the strength of the association between height-for-age z-score and cognitive ability persisted to ages 5-11 years (Sudfeld et al., 2015). Infants discharged from the neonatal intensive care unit who had the poorest head growth from discharge to follow-up, and who were often more critically ill, show the highest risk for cognitive and motor delays at follow-up (Raghuram et al., 2017). Thus, it is important in longitudinal studies of children to track weight, height/length, and head circumference over time to understand nutritional and anthropometric impacts on cognitive, behavioral, and socioemotional development.

#### 4.1.4 | Body composition

Growing evidence that body composition is influenced by chronic stress and adversity further emphasizes the need for validated methods of body composition assessment in the pediatric population. Chronic stress, mediated at least in part through the actions of stress hormones called glucocorticoids, may lead to dehydration, greater fat mass (FM), a loss of muscle mass and strength, and less optimal bone health as described elsewhere (Stefanaki, Pervanidou, Boschiero, & Chrousos, 2018; van der Valk, Savas, & van Rossum, 2018). There is a particularly strong body of literature on the association between chronic stress and centralization of body fat.

For infants specifically, maternal prenatal depression and anxiety are generally found to be associated with impaired offspring growth (Ding et al., 2014; Nasreen, Kabir, Forsell, & Edhborg, 2010). A recent systematic review of the literature showed that children of mothers with postpartum depression have a higher risk of being underweight and stunted in the first year after birth with impaired linear growth after the first year (Farías-Antúnez, Xavier, & Santos, 2018). Various forms of fetal stress may also be associated with later risk for metabolic disease and obesity (Entringer et al., 2012). For example, maternal nutrient restriction leads to increased adipose tissue in the fetus in animal models (Bispham et al., 2003). In humans, growth restriction following stress in utero and in infancy appears to be reversed in childhood and in later life. Exposure to maternal psychosocial stress in utero is associated with higher body mass index (BMI), percent body fat (%BF), insulin resistance, altered lipid profiles, and accelerated cellular aging in offspring in young adulthood (Entringer et al., 2008).

It is thought that most of these associations are due to hyperactivation of the HPA axis and the increase in concentration of stress hormones called glucocorticoids in circulation with central and peripheral effects. High levels of circulating glucocorticoids cause insulin resistance and stimulate adipocyte development (Stefanaki et al., 2018). Hyperactivation of the HPA axis not only affects adipose tissue and insulin receptor-rich tissues directly but also causes changes in eating behavior. Maternal anxiety and depression may, for example, indirectly alter fetal and infant anthropometry and body composition via differences in the mother's dietary intake and dietary quality that she provides for her infant (Farías-Antúnez et al., 2018).

#### 4.1.5 | Obesity

Growing evidence suggests that overnutrition is associated with cognitive and socioemotional functioning as well. Given the high rates of overweight and obesity in the US and globally, understanding how high weight-for-height/length, BMI, and adiposity are associated with developmental outcomes is important for public health. A meta-analysis of 67 studies supports a negative association between obesity and executive functioning, motor skill, attention, and visuospatial performance (Liang et al., 2014). Children with overweight are more likely to show poorer cognitive functioning than their peers with normal weight (Li, Dai, Jackson, & Zhang, 2008). Poorer executive function is associated with behaviors that increase risk for obesity, including greater food intake, less physical activity, and disinhibited eating (Liang et al., 2014). Thus, these associations between obesity and cognition are likely bidirectional. Children with overweight and obesity may also experience social and emotional difficulties in addition to difficulties in certain cognitive domains. Children with overweight are more likely to struggle with social and emotional problems at 6–7 years of age, which worsen for children with severe obesity (Harrist et al., 2016). Individuals experiencing chronic stress may exhibit alterations in stress system functioning, which increases the risk of weight gain and metabolic disturbances (van der Valk et al., 2018). Overweight and obesity are the result of complex etiological processes, including genetics, microbiome, intrauterine environment, food environment, diet, physical activity, and social factors. The association between stress and obesity is also bidirectional (van der Valk et al., 2018). As a result, transdisciplinary research is necessary to understand these complex processes leading to overweight and obesity.

#### 4.2 | Cardiometabolic health and seed science

Organizations such as the American Heart Association (AHA) are increasingly considering the role of early psychosocial factors, including childhood stressors such as maltreatment and poverty, in contributing to health across the lifespan (Suglia et al., 2018). There is a growing understanding that the origins of adult CVD begin at least as early as infancy, and likely during the prenatal period in the case of stress and health problems during gestation. This increasing focus on early psychosocial factors that increase risk for physical health problems creates exciting opportunities for social scientists to collaborate on solving these problems. There is evidence that childhood stress may influence cardiometabolic health through changing the neural circuitry underlying threat detection, reward responsiveness, and fear learning, alterations in the stress and immune systems, and health behaviors, including intake of foods high in sugar or fat or eating in the absence of hunger to regulate negative emotions (Dallman, 2010; Lupien, McEwen, Gunnar, & Heim, 2009; Suglia et al., 2018). SEED scientists are particularly well-suited to assess these mediators and to design interventions that promote health.

Extensive research has demonstrated strong links between physical and mental health problems throughout the lifespan. For example, individuals with CVD and other chronic diseases are more likely to develop mental health problems such as depression and anxiety (Prince et al., 2007). These associations are bidirectional. Those who have depression and anxiety are more likely to adopt negative health behaviors such as smoking, poor diet, sedentary behavior, and social isolation, and to develop substance use disorders (Prince et al., 2007). These behaviors partially mediate associations between mental health problems and CVD, and there is evidence for biological pathways by which mental health may directly influence physical health (Prince et al., 2007). The connection between mental and physical health is especially important for SEED researchers to consider as early stress places children at risk for both mental and physical health problems.

SEED scientists who are interested in associations between physical and mental health increasingly assess measures of health that are "under the skin" in order to better understand common risk pathways to a large number of disorders. For example, within the DOHaD framework, inflammation is a hypothesized mechanism by which stress influences both mental and physical health. Inflammation, a risk factor for CVD, has been tied to poorer cognitive function (Oriá et al., 2016). Recent research has focused on the association between inflammation and depression. For a subset of depressed individuals, inflammation is key to the pathogenesis of depression (Kiecolt-Glaser, Derry, & Fagundes, 2015). This association is especially relevant for individuals with a history of childhood adversity (Baumeister, Akhtar, Ciufolini, Pariante, & Mondelli, 2016; Miller & Cole, 2012). There is now strong evidence that chronic stress, as well as acute stress, results in immunological shifts that persist over time. A meta-analysis across 25 studies shows significant associations of important inflammatory mediators (interleukin-6 [IL-6], tumor necrosis factor-alpha [TNF- $\alpha$ ], and C-reactive protein [CRP]) with self-reported childhood adversity many years before (Baumeister et al., 2016). Interestingly, these effects appear to be magnified by stressful events in later life (Rohleder, 2019) and by obesity (McInnis et al., 2014). Stressors and pathogens may lead to inflammatory responses that are too high or last too long, which can lead to negative health behaviors (e.g., disturbed sleep, fewer social interactions) as pathways to depression (Kiecolt-Glaser et al., 2015). Stress may also lead to negative health behaviors (sedentary behavior, poor diet), which are then associated with increased inflammation and risk for depression (Kiecolt-Glaser et al., 2015). There is some evidence that early stress may amplify crosstalk between the brain and immune systems, increasing risk for depression and cardiometabolic problems (Hostinar, Nusslock, & Miller, 2018). SEED scientists are wellsuited to assess the psychosocial, environmental, and behavioral factors that lead to inflammation and poorer cardiometabolic health as well as understanding how poorer cardiometabolic health may influence mental health and behavior across development. There are many research groups who have been doing excellent work on stress and cardiometabolic risk in populations characterized by high psychosocial risk (e.g., Baldwin et al., 2016; Felder et al., 2020; Lumeng et al., 2014; Miller et al., 2018; Reid et al., 2018; Suglia, Duarte, Chambers, & Boynton-Jarrett, 2012; Winning, Glymour, McCormick, Gilsanz, & Kubzansky, 2016).

### 5 | REVIEW OF METHODS

#### 5.1 | Anthropometric methods

Given the importance of growth status and growth rate as indicators of child well-being and as potential markers of stress, researchers should collect height (or recumbent length for infants up to 24 months) and weight for all participants enrolled in SEED studies. Research assistants should receive initial training and regular review of the appropriate methods for obtaining measurements from qualified anthropometric experts to ensure intra- and inter-rater reliability (see example: De Miguel-Etayo et al., 2014). If conducting a multi-site study, each site should have the same equipment for consistency, and research assistants should be trained to obtain consistent (reliable) and accurate (unbiased) measurements both before the study commences and at intervals while the study is ongoing. Inter-rater reliability is assessed using the inter-rater coefficient of variation (CV) and should be >95% for all measurements. Accuracy of measurement is assessed by comparison of measurements from new research assistants to those with expertise in anthropometric assessment. There should be no difference in the mean values across raters. Training research assistants on properly positioning participants, especially for height and length measurements, and being able to identify landmarks on the body are important for improving accuracy and precision of anthropometry measurements. More detailed training on child growth assessment and use of international reference data is available online through the WHO (World Health Organization, 1995, 2008). A comprehensive set of anthropometric methods published by the Centers for Disease Control (CDC) for use in the U.S. National Health and Nutrition Examination Survey (NHANES) is also available (https:// www.cdc.gov/nchs/data/nhanes/nhanes\_07\_08/manual\_an.pdf). For assessments of anthropometry that are more intrusive (i.e., waist and hip circumference), trained chaperones and/or parents should be in the room for the safety of the participant.

Measuring weight is straightforward, with the greatest challenges including the child holding still long enough for the scale to register a weight, and ensuring that the scale being used is high quality and appropriately calibrated. Measurement of height/length is much more challenging. Small errors in height/length measurement can have substantial implications for understanding growth in the developing child. For example, while an error in measurement of one centimeter (cm) in an average height 20-year-old man, from 177 to 176 cm, changes the height z-score by -0.14 and the height percentile by 5.6%, the same one centimeter error in an average height 3-year-old boy, from 95 to 94 cm, changes the height z-score by -0.26 and the height percentile by 10.1%. The impact of a 1cm error will be even greater at the tails of the distribution: 1 cm error in measurement of a relatively tall 20-year-old man will change the height z-score by 0.14% and 3.07%, while the same 1cm error in a relatively tall 3-yearold boy will change the height z-score by 0.48 and the height percentile 8.2%). The effect of the error is literally exponentiated when using the height to calculate BMI, because the height (the denominator in the BMI equation) is squared in the equation. The precision with which height needs to be measured in children compared to adults is a result of the more constricted range of average height in a 3-year-old (5-95th percentile for height is a 13 cm span, and weight is a 5.5 kg span, and BMI a 3.8 kg/m<sup>2</sup> span) as compared to a 20-yearold (5-95th percentile for height is a 23.5 cm span, and weight is a 40.5 kg span, and BMI an 11.4k g/m<sup>2</sup> span). Stated differently, the difference between normal weight and obesity in a 3-year-old boy is 0.9 kg/m<sup>2</sup>, whereas the difference between normal weight and obesity in a 20-year-old man is 3.6 kg/m<sup>2</sup>. Successful interventions for obesity in children can generally be expected to have an effect size of 0.2 SD units for BMI. Therefore, slight errors in height measurements in children may mask effects of interventions. In addition, for

measurements in adults.

## -Developmental Psychobiology-WILEY

children between 2 and 7 years, it is essential to percentile or *z*-score BMI in particular, as opposed to using raw BMI, because this is the age range in which adiposity rebound occurs. Therefore, the large effects of small differences in height measurement on percentiled and *z*-scored measures are especially impactful in the preschool age range. The same principles apply to measurement of head circumference and waist circumference in children; the constricted range of a child's size leads to the potential for substantial error as compared to

#### 5.1.1 | Measures of linear growth

A stadiometer is used to measure height for anyone 24 months of age or older (Figure 2). These can typically be purchased for \$100-\$200 USD. Ideally, the stadiometer is fixed to a wall without molding in an area with a flat, non-carpeted surface, with a rigid sliding headpiece. Portable stadiometers can be used in field studies, and are equally reliable if assembled correctly, placed on a stable surface, with a headpiece that remains parallel to the floor during measurement. A stadiometer affixed to a beam balance scale is not stable and should not be used. Individuals are measured without shoes. Hats and hair accessories are removed for accurate measurement. Individuals should stand with their backs to the wall and feet flat, together against the wall. Their legs should be straight and shoulders should be level. The head should be positioned in the Frankfort horizontal plane (line of sight is parallel with the ground) and at least one part of their body, such as their buttocks or shoulders, should be touching the wall. The researcher should then lower the sliding horizontal headpiece of the stadiometer until it firmly touches the top of

#### **Child Height Measurement**



FIGURE 2 Child and adolescent height measurement (24 months through adolescence/adulthood). From How to Weigh and Measure Children, by the United Nations Statistical Office. ©1986 United Nations. Reprinted with the permission of the United Nations the head. Height should be read with the researcher positioned at the same level as the headpiece. This process should be done twice, which means the individual should step off the stadiometer and redo the entire measurement procedure. If estimates differ by more than 0.5 cm, a third attempt should be made. The final height should be calculated by taking the mean of the two measurements within the tolerance limit. Incorrect placement of participants against the stadiometer is a common pitfall in height measurement which can lead to substantial error.

For infants less than 24 months of age, length should be measured using a recumbent length board appropriate for the infant's size (Figure 3). Length boards range in cost from \$100–500 USD, depending on size and manufacturer. A thin cloth or tissue paper may be used to make the board more comfortable and to keep the board clean between uses. After removal of clothing, shoes, and any hair accessories, the infant should be placed in a supine position with the crown of the head firmly against the headboard in the Frankfort horizontal plane. Two adults are needed for an infant recumbent length measurement. One (can be the parent) should gently but firmly hold the infant's head in the Frankfort horizontal plane at all times insuring the head remains touching the head board. The second measurer straightens the infant's legs by applying gentle pressure to the knees and holds one foot perpendicular to the footboard. Next, the footboard is moved forward until it rests flat against the sole of at least one of the infant's feet. Measurements should be taken while the infant's shoulders and spine are straight against the length board. At least two measurements should be taken to the nearest 0.1 cm (World Health Organization, 2008). Incorrect placement of infants on the length board is a common pitfall in length measurement.

#### 5.1.2 | Body weight

Weight should be measured with a digital scale or a beam balance scale. Research-grade digital scales can typically be purchased for under \$100 while beam balance scales can range from \$150 to \$400. For all scales, standard weights should be purchased to check the scale for accuracy weekly. If the scale is not measuring the standard weights accurately, it needs to be recalibrated, and if recalibration does not correct the inaccuracy, the scale needs to be replaced. The scale should be placed on firm, non-carpeted flooring. All shoes and heavy clothing must be removed. Children 24 months and older should be directed to stand with feet in the center of the scale. Weight is recorded to the nearest 0.01 kg (e.g., 36.32 kg). As with length, the participant should be measured at least twice, and if the values differ by more than 0.1 kg, the participant should be weighed a third time. The final weight should be calculated as the mean of

#### **Child Length Measurement**



FIGURE 3 Infant length measurement (0 to <24 months). From How to Weigh and Measure Children, by the United Nations Statistical Office. ©1986 United Nations. Reprinted with the permission of the United Nations

the measurements within the tolerance limit. For young children or those unable to stand, it may be appropriate to have a caregiver step on the scale to have their weight documented before holding their child to determine the child's weight by subtracting caregiver weight from total weight. However, it is much more accurate for the child to stand alone on the scale if at all possible.

In infants, weight should be obtained using a calibrated research-grade digital or beam balance scale with a tray large enough to securely hold the infant. After removal of clothing (a clean, dry diaper is permissible), infants should be placed in the center of the scale tray. Researchers should record the measured weight before repositioning the infant and weighing again. The two measurements should have agreement within 0.01 kg. Having a caregiver assist with weight measurements as detailed above for children can also be used for infants who are fussy or have difficulty staying still on the scale for a long enough period of time.

#### 5.1.3 | Head circumference

Head growth is largely completed by 3 years. Head circumference, an important measure of brain growth, is therefore assessed only till 36 months in longitudinal studies. Head circumference is measured using a flexible, inelastic measuring tape between 0.5 and 1 cm wide. Use of an insertion measuring tape (one end inserts into the other) is ideal to prevent having to overlap the tape measure when reading measurements.

Before measurement, any hats or hair accessories should be removed. To measure head circumference, researchers should wrap the measuring tape snugly above the supraorbital ridges of the forehead (typically just above the eyebrows), above the ears, and over the occipital bun, the bony protuberance at the posterior base of the skull to obtain the maximum circumference. Care must be taken that the tape is level on both sides of the head and that the hair is compressed. In contrast to the other measurements, this measure should be taken three times to the nearest 0.1 cm as it is particularly prone to error. The largest measure obtained should be used as the final measure because head circumference is defined as the largest circumference of the head. Appropriate placement of the measuring tape on the head is important for obtaining accurate measurement.

#### 5.1.4 | Waist and hip circumference

Waist circumference is used as a measure of abdominal adiposity. Abdominal obesity is associated with a higher risk of metabolic syndrome (MetS), a cluster of symptoms that predict greater risk for future CVD (He et al., 2015). Waist circumference specifically is associated with cardiovascular risk clustering in children and adolescents (Katzmarzyk et al., 2004). Recent evidence suggests that the waist circumference-to-height ratio may be a reasonable proxy for %BF measurements when gold-standard body fat measurement methods (discussed below) are not available. In a study of children and adolescents, waist circumference-to-height ratio explained 64% of the variance in %BF, which was a greater proportion of the variance than either BMI (32%) or waist circumference (31%) individually (Brambilla, Bedogni, Heo, & Pietrobelli, 2013). Waist circumference is typically not assessed in infants 24 months and younger.

The waist circumference method that has the strongest association with metabolic risk in children is measuring the waist at the narrowest point between the iliac crest (hip bone) and lower costal border (floating rib or bottom end of the rib cage) (Garnett, Baur, & Cowell, 2008: Johnson, Kuk, et al., 2010). To measure waist circumference, a flexible, inelastic measuring tape is used. Small stickers, cosmetic pencils, or washable markers are also helpful. The participant should stand with their feet about hip width apart and gather their shirt slightly above the waist. The research assistant should stand at the participant's right side and either palpate their hip area to locate the top of the hip bone (the iliac crest) or have the participant point out this area themselves. Using a cosmetic pencil or marker, the research assistant draws a small horizontal line right at the top of the hip bone. Alternatively, a sticker may be placed at this location. The same steps should then be performed to locate the bottom edge of the participant's floating rib or rib cage (lower costal border). The research assistant should identify the narrowest point around the waist between these two locations. If there is no clear narrow point, the research assistant should identify the midpoint between these two locations. The research assistant should place one end of the measuring tape at the narrowest point or midpoint and extend the measuring tape around the participant's waist and back to the original location. This is facilitated by having the participant hold the end of the tape at the original location while the research assistant extends the tape. The tape must sit parallel to the floor and be snug but not so snug that it compresses the skin. The measurement should be taken when the participant naturally exhales. The research assistant should read the tape while their eyes are level with the numbers rather than from an angle above or below the numbers in order to obtain an accurate reading. The value should be reported to the nearest 0.1 cm. The measurement should be taken twice, and the average should be used as long as the two values are within 0.5 cm. If not, the procedure should be repeated. The participant should be informed in advance that their waist circumference will be measured so they can dress in clothing that can be lifted to their waist. If the participant is wearing incompatible clothing, the research assistant may offer to have the participant borrow clothing provided by the laboratory to wear for the anthropometry component of data collection. There is evidence that measures of waist circumference in youth with overweight or obesity should include the panniculus (apron of abdominal fat) as it better correlates with absolute body fat measures (Sabin et al., 2014). Waist circumference measurement method should be standardized across all participants for optimal measurement.

While stress researchers should consider inclusion of abdominal or waist circumference measures as indicators of excess visceral adipose tissue deposition in older children and adolescents, these metrics are typically not collected in infants. In one of the few existing infant magnetic resonance imaging (MRI) studies, abdominal visceral adipose tissue comprised 2.4% of all adipose tissue at 2 weeks of age, and <2% of all adipose tissue at 2 months (Gale et al., 2015). There are some infant body composition prediction equations that include other body circumference measures (trunk or thigh, for instance), but by far the more common anthropometric predictors of whole body composition in infancy are weight, length, and subcutaneous skinfold thickness (Demerath & Fields, 2014).

Hip circumference is most often used to calculate the waist-hip ratio, which is used as a measure of cardiovascular risk and a predictor of all-cause mortality in adults (World Health Organization, 2011). For hip circumference, the research assistant stands at the participant's side and locates the widest part of the buttocks. The same flexible, inelastic tape for waist circumference should be used. The research assistant should place one end of the tape at the participant's side at the same height as the widest part of the buttocks. The participant can be instructed to hold the tape at their side and slowly rotate 360° to form a circumference around their buttocks. The tape must be parallel to the ground and snug but should not compress the skin. Hip circumference should be recorded to the nearest 0.1 cm and the average of the two measurements should be used. If the measurements are more than 0.5 cm apart, the procedure should be repeated. Improper measuring tape placement on the waist and hip are common pitfalls for accurately measuring waist and hip circumference. Dividing the waist circumference by the hip circumference is used to determine the waist-hip ratio. Waist-hip ratio may be less useful for assessing abdominal adiposity compared to waist circumference and BMI at least through 17 years of age (Neovius, Linné, & Rossner, 2005).

#### 5.1.5 | Anthropometry and nutrition status

Height (or recumbent length) and weight measurements may be used to track associations between psychosocial stress and growth. A height/length-for-age z-score <-2 using international growth reference charts defines stunting, for example, which is an outcome of chronic malnutrition during childhood, while a weight-for-age zscore <-2 defines wasting, an outcome of acute or chronic malnutrition. Given that weight is partially a function of height/length, body weight is also typically converted to a weight/length or weight/ height index as an indicator of relative weight and of adiposity that essentially removes the effect of height/length. The most common indicator at all ages is BMI, which is calculated as weight (in kg) divided by height (in m) squared. Although BMI can be calculated for infants younger than 24 months, the CDC recommends against using BMI and instead using weight-for-length. Length is especially difficult to accurately measure; thus, squaring the length value as required in calculating BMI will exponentiate errors. In neonates, the ponderal index (weight divided by length cubed) is often used to assess symmetrical versus asymmetrical IUGR. There are also biochemical assessments of nutrition status and dietary intake, but these are not covered in the current review.

#### 5.1.6 | Growth reference charts

Due to the rapidity of growth in infancy, puberty, and throughout childhood, profound changes in weight, height/length, and other anthropometrics occur over short periods of time. In addition, given known sexual dimorphism, with boys being generally taller and heavier than girls, starting before birth, the interpretation of a given anthropometric measure thus depends strongly upon both the exact age at measurement and sex. Therefore, when reporting or analyzing data on height/length, weight, head circumference or derived indicators, in infants, children, and adolescents (birth to age 20 years, when growth is typically completed), age- and sex-standardized percentiles or z-scores should be used, using national or international reference data. The WHO and CDC have published reference data and associated curves for the following growth indicators by sex: weight-for-age, height/length-for-age, weight-for-height/length, BMI, and head circumference-for-age. Height and length have separate reference charts, and the measure should be plotted on the chart reflecting the manner in which the linear growth measurement was obtained, since the same child will be longer when measured lying down (length) as opposed to when measured standing (height). In the United States, the CDC recommends use of the WHO growth curve standards, based on data from healthy, exclusively breast-feeding infants from birth to 24 months of age. There are few differences between the WHO and CDC growth charts from 2 to 20 years of age and either can be used, though the CDC growth charts are commonly used in the United States since they are based on a US population (Centers for Disease Control and Prevention, 2010). Although there are online calculation tools for determining individual z-scores and percentiles relative to the standards, they are inefficient for studies with hundreds or thousands of participants. Instead, published macros may be used with statistical software to compute z-scores and growth percentiles from formatted data files of participant anthropometric data. If chronological age is not available, as may be the case in adoption, or other situations where hospital birth records are not available, age can be estimated using skeletal radiography and other measures of physical maturity. From birth to 24 months of age, the WHO identifies infants below the second percentile on length-for-age as having short stature, below the second percentile on weight-for-length as having low weight-for-length, and above the 98th percentile on weight-for-length as having high weight-for-length. From 2 to 20 years of age, the CDC identifies stature-for-age below the 5th percentile as short stature, BMI-for-age below the 5th percentile as underweight, BMI-for-age ≥85th and <95th percentile as overweight, and BMI-for-age ≥95th percentile as obesity. In addition, growth measures that increase or decrease more than two major centile bands indicate either rapid or slow growth, especially if the pattern persists over time, which indicates that nutritional assessment is needed. Tracking along a proximate centile band within 2 SD of the median generally indicates healthy growth (World Health Organization, 2006). Specific growth reference charts are available for premature infants prior to reaching 40 weeks gestational age, as well as for children with certain genetic conditions, since normal

growth patterns in these populations differ. For children who were born prematurely, growth is charted correcting for the number of months premature for at least the first year after birth.

#### 5.2 | Pubertal assessment

Pubertal growth trajectories, especially early pubertal onset, are associated with increased risk for adult obesity and CVD in both female (He et al., 2010; Lakshman et al., 2008, 2009) and male youth (Widén et al., 2012). Puberty is a process that develops from complex and coordinated neuroendocrine changes that lead to changes in sexual characteristics and eventually results in the capacity for reproduction. Puberty is a dramatic transition physically, emotionally, cognitively, and behaviorally in adolescent populations (Dorn, 2006). Appropriate measurement and understanding of the physiology of puberty and its impact on physical and mental health would both enhance knowledge and could improve outcomes for adolescents.

As the details of the neuroendocrine aspects of puberty and its components are beyond the scope of this review, we recommend Dorn (Dorn & Biro, 2011) and Mendle (Mendle, Beltz, Carter, & Dorn, 2019) for excellent overviews. Generally, puberty consists of two distinct and overlapping processes: adrenarche and gonadarche. Adrenarche begins around age 6-8 years (Byrne et al., 2017; Lee & Styne, 2013; Styne & Grumbach, 2016) with the maturation of the adrenal gland and a steep rise in adrenal steroid hormones. Gonadarche follows adrenarche approximately 1-2 years later. Reliant on the coordinated endocrine function of the hypothalamus, pituitary gland, and gonads (hypothalamic-pituitary-gonadal axis), gonadarche is a gradual process that lasts for about 4-5 years (Bordini & Rosenfield, 2011; Rey, Campo, Ropelato, & Bergadá, 2016; Styne & Grumbach, 2016). There are sex differences in gonadarche and adrenarche onset: it begins approximately 1-1.5 years later for males compared to females. Menarche, or a girl's first menstrual cycle, is a relatively late gonadal event, occurring between 1.5 and 3 years after breast development (thelarche) begins. The associated changes with gonadarche are considered "central" pubertal development, and hypotheses regarding peripheral and central pubertal development will require different focal variables for pubertal assessment.

There are a number of ways to assess pubertal status. The "gold-standard" method is Tanner staging by a trained assessor's examination (Dorn & Biro, 2011). Tanner staging consists of five stages of pubertal development, and categorizes pubertal status based on pubic hair development, as well as breast development in girls, and testicle development in boys (Marshall & Tanner, 1969, 1970; Tanner, 1962). Tanner assessments can be completed by nurses or medical personnel who are trained in the specific Tanner physical exam; reliability across assessors should be routinely checked over the course of a study. Tanner Stage 1 refers to a pre-pubertal stage and is defined by no visible signs of development in the domains of breast, testicle, or pubic hair development. Tanner Stage 5 refers to "complete" pubertal development or full physical maturation. Physical examination, not just visual examination, should be done to ensure accuracy in Tanner staging given that breast and adipose tissue cannot be differentiated visually, and testicular size cannot be evaluated without palpation (Dorn & Biro, 2011). Visual examination for Tanner staging is less accurate, though still acceptable if physical examination is not possible (Dorn & Biro, 2011). Of note, systematic error in measuring breast development will be introduced by visual examination alone, with obese girls erroneously identified as having more advanced breast development because adipose tissue is not visually distinguishable from breast tissue in the developing breast; this distinction can only be made by palpation. Thus, visual rating of breast development will erroneously identify girls with obesity, compared to those without, as having more advanced pubertal development. In either the case of visual examination or physical examination with palpation, researchers should take great care in ensuring a safe examination environment for participants, reviewed in detail in Dorn (2006). In addition to a physical examination, studies have also collected blood for serum pubertal hormone concentrations in concert with a physical examination (Brooks-Gunn & Warren, 1989; Susman et al., 1987). For more detail on serum pubertal hormone concentrations, we refer the reader to Dorn (2006).

There are also self-report methods of pubertal staging, however, there are very few recent studies that compare inter-rater agreement between self-report and physician-assessed pubertal status (Dorn & Biro, 2011). For Tanner staging, parents or youth can use either photographs or line drawings for parent- or self-assessment of which stage the youth participant falls within (Biro & Dorn, 2005; Morris & Udry, 1980). Line drawings, designed by Morris and Udry, are less graphic and are an acceptable, though lower quality, approach than photos if researchers and participants prefer a less realistic depiction of the developing body. Another self-report measure of pubertal development, the Petersen Pubertal Development Scale (PDS) (Petersen, Crockett, Richards, & Boxer, 1988), focuses on secondary sex characteristic development, including body hair, skin changes, and linear growth acceleration (i.e., a growth spurt) for both sexes; facial hair and vocal changes in males; and menarche and breast development for girls. The PDS asks participants to rank themselves compared to their same-age peers on each dimension, and it is therefore not anchored in specific physical changes but rather the participants' perceptions of their own development relative to others (Mendle et al., 2019). The advantage of the PDS is that it is the most widely used self-report measure of puberty and its psychometric properties are well-documented (Petersen et al., 1988). A limitation of the PDS is that pubertal staging is weighted more heavily toward changes that become evident in mid- to late puberty such as facial hair, vocal changes, and menarche, and less heavily toward the changes that are evident in early puberty when assessed with Tanner staging. There is evidence that the correlation of salivary hormone levels with the PDS is similar in magnitude to the correlation with physical examination (Shirtcliff, Dahl, & Pollak, 2009).

Regardless of the method used, investigators should seek appropriate collaborators with expertise in both pubertal development and the assessment method of interest. Measures selected should be theoretically relevant to the research question and researchers should establish a rationale about which measure to use for appropriate hypotheses. In addition to the scientific integrity of selecting a measure of puberty for a study, there are multiple methodological strategies to consider when working across diverse social contexts. These contexts include populations of racially and ethnically diverse youth, low-income youth, and youth with a history of life stress or trauma. More detail is provided by Mendle (Mendle et al., 2019).

One dimension of risk in pubertal development is premature adrenarche, which is puberty onset that occurs in girls aged 8 years or younger, or in boys aged 9 years or younger (Siegel, Finegold, Urban, McVie, & Lee, 1992). Girls with premature adrenarche have been demonstrated to have higher risk of metabolic syndrome, mood disturbances, and behavioral problems (Dorn & Biro, 2011). Similarly, both early maturing and later maturing boys show greater mood and adjustment problems (Mendle & Ferrero, 2012). There has been more research recently on trajectories of pubertal development, including measures of pubertal tempo, which assess how quickly or slowly children or adolescents proceed through puberty. Models using longitudinal data can be used to estimate the rate of change in pubertal stages per year or estimating when the peak rate of change occurs (Beltz, Corley, Bricker, Wadsworth, & Berenbaum, 2014). Faster pubertal tempo may be associated with steeper declines in reports of parent-child closeness across puberty (Marceau, Ram, & Susman, 2015) and, for girls, greater internalizing and externalizing problems (Marceau, Ram, Houts, Grimm, & Susman, 2011). Peak height velocity may be another measure of interest to researchers, and there are data demonstrating associations between pubertal timing and peak height velocity for boys and girls (Granados, Gebremariam, & Lee, 2015). There is not yet enough research on pubertal tempo or peak height velocity to understand a risk level that reliably predicts increased risk for cardiometabolic health or psychosocial functioning.

#### 5.3 | Body composition

Although BMI tends to be correlated with total adiposity, it is a poor predictor of pediatric adiposity at the individual level in both term (Bell et al., 2018) and preterm (Ramel, Zhang, Misra, Anderson, & Demerath, 2017) infants, and is insufficient for identifying over a quarter of children with high body fat percentage (Javed et al., 2015). In addition, BMI and other simple anthropometric indicators do not provide a measure of lean versus adipose tissue accrual, or the regional distribution of adipose tissue accrual (e.g., visceral vs. elsewhere), which respond differently to chronic stress (Stefanaki et al., 2018). Glucocorticoids cause insulin resistance and have a stimulatory effect on the maturation of preadipocytes, undifferentiated cells that can be stimulated to form a fat cell (Stefanaki et al., 2018), and appear to cause a redistribution of white adipose (fat) tissue toward the abdominal region (van der Valk et al., 2018). Therefore, use of body composition techniques to estimate total lean and adipose tissue volumes as well as those that provide regional estimates of adiposity may be considered, depending on the study question.

Body composition analysis can provide a more specific assessment of %BF and lean tissue, and can be used from infancy through adolescence. Field methods for body composition assessment are typically two- or three-component models, dividing the body into FM and fat-free mass (FFM) or FM and the FFM components of total body water and dry FFM (Ellis, 2000; Weber, Leonard, & Zemel, 2012). Methods reviewed below are generally feasible in infancy as well as at older ages (Demerath & Fields, 2014).

The methods described below range from easy and inexpensive to complex and expensive, with increasing expense typically associated with greater specificity of body composition components and greater accuracy in assessment of whole-body FM and FFM. Body composition assessment from 0 to 5 years is particularly difficult, with major limitations to each method. A recent workshop at the National Institute of Diabetes and Digestive and Kidney Diseases concluded that there are no approaches available to track the natural history of body composition from birth to adolescence and adulthood (Gallagher et al., 2020). There is also debate regarding the level of body fat that confers additional health risks. It has been proposed that, similar to BMI percentiles by the CDC, youth above the 85 and 95th centiles in %BF should be considered as "overfat" and "obese" (McCarthy, Cole, Fry, Jebb, & Prentice, 2006). However, absent data regarding the threshold at which additional health risk occurs, there is no consensus on what level should be considered high risk (Laurson, Eisenmann, & Welk, 2011). Therefore, it may be useful to examine %BF continuously until risk levels are determined.

#### 5.3.1 | Skinfold measurements

Measuring skinfold thickness (SFT) is an inexpensive method to assess the depth of subcutaneous fat tissue and can also be used with published prediction equations to estimate total body adiposity and %BF. Skinfold measurements are taken using calipers applied to different locations of the body. Generally, both central or trunk sites (e.g., subscapular, abdominal, and suprailiac skinfolds), and peripheral or limb sites (e.g., biceps, triceps, and calf skinfolds) are obtained. Skinfold measurements are taken to the nearest 0.1 mm. The resulting measurements may be used on their own or in published prediction equations (Demerath & Fields, 2014) to estimate body fat percentage (e.g., Slaughter et al., 1988). Advantages of this method are that it is inexpensive, portable, and can be completed in less than 10 min per participant. Disadvantages are that because of the importance of placement of the calipers it can be difficult to obtain measurements with high inter-rater reliability, and measurements are less accurate in children and adolescents with obesity compared to those without obesity (Watts et al., 2006). Reference curves by age and sex have recently been developed for children and adolescents to compare adiposity to US samples (Addo & Himes, 2010; Laurson et al., 2011), similar to BMI growth curves by the CDC and WHO. However, these reference curves are based solely on US children and

may not be valid for SEED researchers studying non-US populations. Skinfold measurement may be the best choice for researchers who want a cost-effective measurement and have the resources to train and retrain research assistants to obtain reliable measurements. In infants, SFT can be affected by fluid status, type of caliper used, and the length of time the caliper is applied to the skin (Heyward & Wagner, 2004; Strydom, Van Niekerk, & Dhansay, 2017). There are a number of prediction equations for estimation of %BF from SFT in infants that have moderately high validity ( $R^2 > .90$ ) but few have been successfully cross validated in independent samples (Demerath & Fields, 2014). While several prediction equations have been proposed, nearly all yield biased measures of FM or %BF (Catalano, Thomas, Avallone, & Amini, 1995; Dejerlein, Thornton, Hull, Paley, & Gallagher, 2012; Lingwood et al., 2012). Particularly at or near birth, body weight and length alone or together explain a very large proportion of the variance in FM and/or FFM; the addition of more error-prone SFT tend to increase validity only slightly. Nonetheless, SFT may be useful in the field for within-sample estimates of regional and total adiposity as long as researchers are aware of the limitations and biases of their chosen prediction equations. In a large study of 8- to 19-year-old NHANES participants, SFT measurements were compared with BMI, using dual-energy X-ray absorptiometry (DXA) as the gold standard. They found that SFT and BMI were both fairly accurate for determining excess body fatness. However, for children with higher levels of adiposity, BMI was more strongly associated with DXA-calculated body fatness than the sum of the triceps and subscapular SFT. However, for children with low body fatness, SFT measurement was more accurate than BMI (Freedman, Ogden, Blanck, Borrud, & Dietz, 2013). Researchers traditionally obtain SFT measures as a supplement to BMI measurement, and not in isolation.

#### 5.3.2 | Bioelectrical impedance analysis (BIA)

BIA measures body composition indirectly by sending a weak electrical current through the body and measuring resistance. Body tissue with low fluid content, such as adipose tissue, produces more resistance than lean tissue with high fluid content. Published prediction equations may then be used to calculate %BF and FFM. BIA devices can be single-frequency (SF-BIA), multi-frequency (MF-BIA), or spectroscopy devices (BIS) (Mulasi, Kuchnia, Cole, & Earthman, 2015). In general, spectroscopy devices, which measure impedance over a wide range of frequencies, may be more optimal for use in pediatric populations because they do not rely on factors such as age, sex, and race/ethnicity to calculate estimates of body composition (Deurenberg & Deurenberg-Yap, 2003; Mulasi et al., 2015). Many spectroscopy devices include a stand-on scale, are easy to use, and are often portable. BIA is less expensive than methods like DXA and air displacement plethysmography but still more expensive than height, weight, waist circumference, and skinfold measures. A recent systematic review reports that BIA is highly correlated with DXA, but tends to underestimate FM in children and adolescents (Chula de Castro et al., 2018). MF-BIA is precise enough to be widely

used in epidemiological studies with children (Tompuri et al., 2015). More work is needed to increase the accuracy of SF-BIA and MF-BIA equations for use in children of different races/ethnicities and ages as differences in body density can introduce error. However, BIA may be especially useful for researchers who are seeking a method apart from BMI alone, that is also convenient, relatively easy to use, and a reasonably accurate measure of body composition.

In infants, the use of SF-BIA and MF-BIA, which rely on stable hydration status, is challenging due to changes in total body water that occur throughout infancy (Fomon, Haschke, Ziegler, & Nelson, 1982). The accuracy of BIS data (particularly measurement of extracellular resistance) is affected by infant movement and dietary intake, which is important to consider before use of this method (Sesmero, Mazariegos, Pedrón, Jones, & Solomons, 2005). Furthermore, a validated prediction equation for %BF does not yet exist for any type of bioimpedance analysis in infants. Due to the rapidly changing picture of intracellular and extracellular water volumes, and changes in lean mass density in the first months of life, BIA is not often used for infant body composition assessment.

### 5.3.3 | Dual-energy X-ray absorptiometry (DXA)

Originally used to study bone demineralization and osteoporosis, Dual-energy X-ray Absorptiometry (DXA) is now considered a "gold standard" in clinical body fat assessment as it can precisely differentiate lean and fat tissues (Cornier et al., 2011). DXA is considered a gold standard because it directly measures body fat rather than using indirect measures of body fat such as BMI (Banack, Wactawski-Wende, Hovey, & Stokes, 2018) and provides estimates that are highly correlated with other direct but more impractical measures such as computerized tomography (CT), which can be more expensive, time-consuming, and lead to more radiation exposure (Direk et al., 2013; Snijder et al., 2002). Even though DXA machines are expensive, they are often available within obesity treatment clinics and in university-affiliated academic medical centers. Clinicians and researchers in these settings may be open to collaborating with outside researchers. Researchers may be able to rent use of the equipment or pay per scan rather than investing in DXA equipment.

DXA uses X-rays generated at two different energies and calculates tissue composition using differential attenuation of radiation by tissues of different densities. The two main commercial manufacturers of DXA instruments are GE-Lunar and Hologic (Laskey, 1996). DXA can assess regional body fat distribution such as abdominal fat. DXA does not differentiate between visceral fat and subcutaneous fat directly. This value is therefore calculated by a technician using the DXA software and an algorithm for calculating visceral fat (Kaul et al., 2012). The calculated value is both accurate and valid in multiethnic cohorts of adults (Micklesfield, Goedecke, Punyanitya, Wilson, & Kelly, 2012; Neeland, Grundy, Li, Adams-Huet, & Vega, 2016). It has also been used successfully in pediatric populations with highest accuracy in children who are overweight and obese (Bosch et al., 2015). Recently, the algorithm has become

## WILEY-Developmental Psychobiology

available in both the GE-Lunar and Hologic DXA software for the calculation of visceral adipose tissue. At the tail ends of the body fat distribution in children, DXA may underestimate %BF in a child with low %BF and overestimate BF% in a child with high BF% (Sopher et al., 2004). Working with a DXA technician or expert who can appropriately calculate this measurement for children is important.

Assessment of body fat by DXA requires about 20 min for a full body scan and involves very little radiation. The radiation exposure is less than one would receive from a standard chest X-ray or a dental X-ray. It is, therefore, appropriate and acceptable for children and for repeated measures (Plank, 2005). The scan requires the participant to lie still on their back for approximately 15 min while an X-ray is taken of their entire body. Clothing must be free of metal components (e.g., underwire, buttons, or snaps). A hospital gown may be provided. Caregivers can remain in the room while the scan takes place. Participants must hold still for the duration of the scan. which may be challenging for younger children or children with developmental or behavioral challenges. Additionally, female participants (post-menarche) are asked for the date of their last menstrual period and must submit a urine sample for a pregnancy test before scanning, given the risk of radiation exposure during pregnancy to the fetus. Researchers must develop a protocol to address issues of an unexpected positive pregnancy test in a minor as part of the research process. One pitfall related to DXA is the need to calibrate the machine each day before a scan and to schedule a major recalibration of the machine periodically.

A limited number of studies examining DXA and infant body composition have been conducted, most likely because DXA involves exposure to very low but measurable doses of ionizing radiation. For this reason, use of DXA for longitudinal monitoring of body composition in infants is not often performed (Demerath & Fields, 2014). Nonetheless, DXA is safe, relatively fast (about 5 min of scan time), and well-tolerated by most infants. DXA has unique value for body composition assessment because it provides not only total but also regional (trunk vs. limb) estimates of fat, bone, and lean tissue mass. This is very useful when one is concerned with differential impact of early exposures on the growth of different organ systems (namely the musculoskeletal vs. the adipose tissue organs) and central versus peripheral fat deposition. DXA measurements can be affected by movement of the infant, which may lead to missing data from unreadable scans, and overestimation of both FM and FFM (Koo, Massom, & Walters, 1995). There are methods, however, to compensate for body movement using advanced DXA analysis techniques, including "limb reflection"-replacement of limb data for the limb with movement artifact with data from the opposing stationary limb (Shepherd, Ng, Sommer, & Heymsfield, 2017). Infants can be swaddled (while attending to American Academy of Pediatrics [AAP] guidelines regarding swaddling) and watch a video screen during the scan, both of which result in low movement of the body and head. One notable issue with DXA is that different software versions and different machines may yield systematic differences in body composition results for the same participant, necessitating cross-validation using a phantom. In infants, FM values tend to be higher using



**FIGURE 4** Bod Pod<sup>™</sup> with pediatric option. Photo used with permission from COSMED USA, Inc

DXA as compared to air displacement plethysmography (Fields, Demerath, Pietrobelli, & Chandler-Laney, 2012).

# 5.3.4 | Air displacement plethysmography (BOD POD<sup>™</sup>/Pea Pod<sup>™</sup>)

Air displacement plethysmography (ADP) was first introduced in 1995 and has become more popular in recent years due to its ease of administration and high accuracy. It has notable strengths for research in children due to its non-invasive nature. The most popular commercial example is the BOD POD<sup>™</sup>, which calculates body volume using air pressure differences in the chamber of the machine with and without the individual being measured. The BOD POD<sup>™</sup> is highly accurate (Fields & Goran, 2000) but tends to yield higher values of %BF than DXA (Vicente-Rodríguez et al., 2012). The BOD  $\mathsf{POD}^{\mathbb{M}}$  can be used from children aged 2 years through adulthood. Children must be a minimum of 10 kg for body composition assessment in the BOD POD<sup>™</sup>. Individuals who are age 7 years and older sit directly in the seat in the chamber while children aged 2-6 years sit in a high-chair attachment inside the chamber. Individuals are required to wear clothing that is skin-tight, such as swimwear and swim caps. Children typically enjoy the BOD POD<sup>™</sup> because it resembles a space ship, and they are able to watch a movie during the approximately 5-min measurement time (Figure 4). Body composition

results are less accurate when the child moves or sneezes inside the chamber during the test, which makes use of the BOD POD<sup>™</sup> more difficult in younger children and those who would be behaviorally challenged by sitting still in an unfamiliar chamber, wearing unfamiliar clothing, and separated from the parent. Pressure changes due to respiration in the BOD  $POD^{\mathsf{M}}$  are a source of error, best accounted for by simultaneous collection of lung volume. As this is difficult for children to perform, predicted values can be used instead. It must be noted that in SEED research, children who have experienced high levels of psychosocial stress are more likely to have problems with emotion and behavior regulation (Shonkoff et al., 2012), which can make sitting still for 5 min very difficult. If only children who can sit still are included in a study, these samples are likely biased. Thus, this movement issue can be scientifically problematic, especially among children younger than age five or children with behavior regulation difficulties.

Similar to DXA, BOD PODs<sup> $\sim$ </sup> are expensive but are becoming more common in research centers on many campuses as well as in sports facilities. It may be possible to rent use of the BOD POD<sup> $\sim$ </sup> or pay per scan instead of purchasing a machine. The BOD POD<sup> $\sim$ </sup> provides highly accurate body composition assessments that are non-invasive and easy to conduct, but its use is limited by requirement that the participant be able to sit still for 5 min.

The Pea Pod<sup>™</sup> (Cosmed, Inc.), is currently the only available device to conduct ADP measurements in infants weighing between 1 and 8 kg (Figure 5). To determine body composition components, the Pea Pod<sup>™</sup> utilizes the infant's weight and length in addition to gas laws and Fomon's (Fomon et al., 1982) or Butte's (Butte, Hopkinson, Wong, Smith, & Ellis, 2000) age- and sex-specific densities for infant FFM to estimate total body volume, which, along with body mass, yields total body density (Fomon & Nelson, 2002; Ma et al., 2004). Prediction equations then use total body density to calculate FM, FFM, and %BF. Moderate movement and crying do not influence Pea Pod<sup>™</sup> measurements of body composition, making it ideal for use in infants weighing less than 8 kg (Demerath & Fields, 2014; Ellis et al., 2007; Ma et al., 2004). ADP has been validated against the four-component model in infants, yielding a slightly but not significantly higher estimate of %BF than the four-component model (Ellis et al., 2007). Due to its reliability, precision, and accuracy in determining %BF, ADP is recognized as a validated method for assessing body composition in both preterm and term infants (Ellis et al., 2007). Among infants with growth patterns within the normal range, the upper weight limit (8 kg) of the Pea Pod<sup>™</sup> can be reached as young as 3 months of age or as old as 10 months of age; half of normally growing infants reach 8 kg by age 6 months. Infants who are heavier for age will therefore be unable to participate in the Pea Pod<sup>™</sup> measurement, which can introduce systematic bias. Age 3 months is the last age at which almost all infants in a cohort will be able to reliably fit in the Pea  $\mathsf{Pod}^{\mathsf{M}}$ . Beyond this age, infants at the heavier end of the growth curve will be systematically excluded from measurement. Use of the BOD POD<sup>™</sup> or Pea Pod<sup>™</sup> between ages 6 months and 2-3 years (or whenever the child can sit still for 5 min), remains an unresolved challenge.



**FIGURE 5** Pea Pod<sup>™</sup> body composition system for infants. Photos used with permission from COSMED USA, Inc

Although the BOD POD<sup>™</sup> and Pea Pod<sup>™</sup> are user-friendly, it is important that the research assistant properly calibrates the machine prior to assessing a participant. In addition, it is important to properly prepare participants with swimwear and swim caps or other tight clothing so that the volume measures are accurate. Failing to provide participants with proper clothing or to calibrate the machine are two common pitfalls that can result in an inaccurate measurement.

#### 5.4 | Cardiometabolic risk

#### 5.4.1 | Blood pressure

The AAP and the AHA recommend routine blood pressure measurement starting around age 3 years (Flynn et al., 2017). Two types of blood pressure are measured: systolic and diastolic. Measures are presented with the systolic blood pressure as the numerator and diastolic blood pressure as the denominator. Systolic blood pressure assesses the pressure in the blood vessels at the moment the heart muscle contracts. Diastolic blood pressure measures the pressure in the vessels while the heart is at rest between contractions. Systolic and diastolic blood pressure are measured in millimeters of mercury (mmHg) and are typically highly correlated. Systolic and diastolic blood pressure values can be used as continuous measures. Percentiles for pediatric populations based on age, sex, and height are derived from studies of approximately 50,000 children and adolescents (Flynn et al., 2017). In the pediatric population, it is essential to convert raw blood pressure measurements into percentiles based on age, sex, and height. For example, a blood pressure of 100/60 is the 95th percentile in a relatively short 3-year-old boy, the 50th percentile in a relatively tall 7-year-old boy, and well below the 50th percentile for an adolescent. Values can also be used to classify children into established pediatric risk groups: normal blood pressure, elevated blood pressure (>90th percentile), Stage 1 hypertension (≥95th percentile), and Stage 2 hypertension (≥95th percentile + mmHg) (Flynn et al., 2017). These cutoffs are statistical as there is yet no established blood pressure level during childhood and adolescence that leads to poor cardiovascular outcomes in adulthood (Flynn et al., 2017). These cutoffs have clinical importance, as treatment of sustained hypertension in youth is necessary to prevent or reverse organ damage from sustained high blood pressure (Flynn et al., 2017).

There is strong evidence that blood pressure tracks from childhood to adulthood in diverse populations (Chen & Wang, 2008). However, blood pressure as well as obesity and serum cholesterol or low-density lipoprotein (LDL) do not track (Berenson & Bogalusa Heart Study Research Group, 2002). Nonetheless, childhood blood pressure measures may predict who is likely to develop high blood pressure later in life and who may benefit from early intervention. Indeed, high blood pressure above certain cutoffs starting at age 5 years predicts metabolic syndrome in adulthood, which is a cluster of symptoms that predicts cardiovascular risk later (Sun et al., 2007).

Blood pressure is most accurately measured using the auscultation method, in which a trained individual listens for the appearance and disappearance of a tapping sound (Korotkov sounds) through a stethoscope placed on the arm as a blood pressure cuff is slowly deflated. The pressure of the cuff at which the sound is first heard is the systolic blood pressure, and the pressure of the cuff when the sound disappears is the diastolic blood pressure. Blood pressure measured via automatic devices correlates reasonably well with auscultation and has the advantages of ease of use and limiting reader error (Urbina et al., 2008). There are many commercially available blood pressure monitors, but not all have been validated in children. There is a website that compiles independent testing results and lists monitors which have passed a national standard of performance (www.dableducational.org) (Urbina et al., 2008). Monitors are rated as: recommended, questionable, or not recommended.

It is important to use the correct size blood pressure cuffs to obtain an accurate blood pressure measurement. The cuff width should be at least 40% of the mid-upper arm circumference (Urbina et al., 2008). Child-sized blood pressure cuffs can be purchased to accommodate smaller arm width. Another important consideration is to measure blood pressure *at rest* (Urbina et al., 2008). If a child has been active, it is important to have the child sit and relax for at least 5–10 min before blood pressure measurement, with activities including playing a quiet, seated game or watching a calming video. Sitting and relaxing may be difficult for children with behavior regulation

or anxiety problems, issues which disproportionately affect children who have experienced high levels of psychosocial stress.

### 5.4.2 | Arterial stiffness

The SphygmoCor system is a clinical and non-invasive measure of arterial stiffness. Arterial stiffness is indicated by the radial and carotid artery augmentation index (Aix; corrected to a heart rate of 75 bpm) and pulse wave velocity (PWV). The augmentation index measures the relative magnitude of the reflected (i.e., retrograde) pulse wave early in the cardiac cycle, with greater values indicating increased arterial stiffening (Kelly et al., 2014). Pulse wave transit time increases within stiffer arteries and is manifested by higher pulse wave velocity values (Kelly et al., 2014). As for the test itself, a small device that looks like a pen is gently placed on the participant's neck, wrist, and foot to measure blood flow. The procedure is painless but reguires the participant to lie still in a bed for 20-30 min, which can be challenging for some young participants, especially participants who have experienced high levels of psychosocial stress. We direct the reader to an excellent overview by Savant and colleagues (Savant, Furth, & Meyers, 2014) for measuring arterial stiffness in pediatric populations, with considerations that extend to the entire procedure. It is important to note that dedicating 20-30 min may not be possible for researchers trying to minimize participant burden if arterial stiffness is not a central measure.

#### 5.4.3 | Carotid intima media thickness (cIMT)

cIMT assesses the progression of atherosclerosis (plaque build-up) in the carotid artery, which is a predictor of CVD. This measure assesses thickness of the two layers of the carotid artery, with greater thickness indicating greater disease progression even when individuals are asymptomatic. Children and adolescents aged 6–18 years with hypertension, elevated BMI, and high cardiovascular reactivity show elevated cIMT (Baroncini, Sylvestre, Baroncini, & Pecoits Filho, 2017; Doyon et al., 2013; Lambiase, Dorn, & Roemmich, 2012). High cumulative psychosocial risk, measured prospectively in early life, is associated with elevated cIMT in young adulthood even after adjusting for adult SES, health behavior, and depressive symptoms (Hakulinen et al., 2016). There is evidence in adolescents aged 14–16 that higher life stress is associated with heightened cardiovascular reactivity which then predicts elevated cIMT (Low, Salomon, & Matthews, 2009).

cIMT is more difficult to assess than many of the measures reviewed here. It requires the use of ultrasound and technicians with proficiency assessing cIMT. Thus, this method is most suitable for use in collaboration with teams trained to collect valid and reliable assessments of cIMT. In addition, atherosclerotic processes such as increased cIMT are more likely to be detectable during adulthood when individuals are most likely to develop CVD. Thus, this measure currently has less utility in infants and children than in adolescents and adults, particularly when investigating associations with psychosocial stress as opposed to traditional cardiovascular risk factors like hypertension. However, autopsy studies have demonstrated that even young children can develop plaque in their arteries (Berenson et al., 1998), suggesting cardiovascular risk is present as early as childhood. Investigations to date may be underpowered to detect associations between psychosocial stress and cIMT in children. A strength of using this measure-besides its utility in predicting cardiovascular risk-is that reference values have been established in children and adolescents to be able to compare cIMT across populations (Doyon et al., 2013). However, there are some pitfalls of using the measure, including difficulty with measuring certain carotid segments and errors from noise in the image due to refraction. Software should allow for manual review and correction of edge detection in order to fix these errors. Overall, this measure is unlikely to be easily added to studies of children by SEED researchers unless it is a main focus of the research, though it is an important measure to be aware of, particularly for SEED researchers studying adolescent and adult populations.

# 5.4.4 | Selected serum measures of cardiometabolic health

There are a number of guidelines regarding how to obtain blood samples in pediatric populations, and we refer the reader to guidelines on blood draws from the WHO (World Health Organization, 2010). It is especially helpful to work with phlebotomists who have experience with pediatric populations both for the comfort of the child and for the ease of the blood draw. A recent study found that almost half of phlebotomists surveyed lacked training in child development (Piazza et al., 2018), which can make it difficult to obtain a sample successfully. That same study found that the primary challenge to pediatric blood draws was anxiety in children and their parents (Piazza et al., 2018). When conducting blood draws, it is best to schedule them in the morning as they often require fasting (e.g., 8 hr, water only). Even pre-operatively, children are only asked to not eat solids for 6 hr, liquids like milk for 4 hr, and clear liquids like juice for 2 hr prior to surgery. Juice consumption will alter the results of a blood draw intending to obtain fasting blood glucose, for example. It is not ethical to require a child to avoid eating or drinking for more than 2 hr while awake, and prolonged fasting of more than 6 hr while awake can have adverse physiological consequences (Andersson, Schmitz, & Frykholm, 2018). Blood draws on children that must be fasting, obtained only for research purposes, therefore almost always must be obtained first thing in the morning, soon after the child awakens. Samples can be sent to a medical center or diagnostic laboratory for analysis, and while they can be kept on ice for a few hours, the blood needs to be processed that day. Laboratory staff can provide guidance on blood volumes needed for each test and any sample processing instructions. There are limits to the amount of blood that can be safely drawn per day that vary based on the child's age. For example, the maximum safe blood draw for an

average-sized 3-year-old is about 25 ml (five teaspoons), whereas for the average-sized 6-year-old, the maximum amount is about 35 ml (seven teaspoons).

Fasting plasma lipids are commonly measured cardiometabolic risk factors in adults. These include (fasted) measurements of total cholesterol, low density lipoprotein-cholesterol (LDL-C), high density lipoprotein-cholesterol (HDL-C), and triglycerides for lipid levels in addition to glucose and insulin levels. Two types of dyslipidemia (abnormal amounts of lipids in the blood) place children at risk: (a) elevated LDL-C levels and (b) a combination of moderate to severely elevated triglycerides, normal to mildly raised LDL-C levels, and low HDL-C levels (Expert Panel on Integrated Guidelines & Risk Reduction in Children, 2011). Universal screening for dyslipidemia is recommended for children between 9 and 11 years and adolescents between 18 and 21 years of age (Expert Panel on Integrated Guidelines & Risk Reduction in Children, 2011). The guidelines, published in 2011 by the National Heart Lung and Blood Institute and endorsed by the AAP, recommend a single fasted blood draw. While universal screening is recommended from 9 to 11 and 18 to 21 years, selective screening for children with risk factors is recommended from 2 to 21 years.

Elevated fasting insulin levels, a marker of insulin resistance, are associated with atherosclerosis and cardiovascular morbidity (Després et al., 1996; Ruige et al., 1998). In children, fasting insulin levels predict later blood pressure (Taittonen et al., 1996), weight gain (Odeleye, de Courten, Pettitt, & Ravussin, 1997), and cardiovascular risk factors (Bao, Srinivasan, & Berenson, 1996). Two methods for assessing insulin levels include the oral glucose tolerance test and measurement of fasting insulin. In addition to the single measure of insulin, plasma glucose can be measured with the same blood draw in order to calculate the homeostasis model assessment-estimated insulin resistance (HOMA-IR) score from a standard equation that uses single measurements from fasting plasma glucose and insulin levels (Levy, Morris, Hammersley, & Turner, 1999). HOMA-IR assesses insulin resistance, a precursor for Type 2 diabetes, and is especially useful in epidemiologic and population-based studies.

There is evidence that psychosocial stress during childhood is associated with small shifts toward greater inflammation already during childhood (Kuhlman, Horn, Chiang, & Bower, 2019), though the studies to date have been small and heterogenous. Studies incorporating systemic inflammation or in vivo assessment of immune cells into SEED research will aid our understanding of how stress gets under the skin to influence health. Commonly used serum measures of cardiometabolic risk include inflammatory proteins such as CRP, IL-6, and TNF- $\alpha$ . CRP, IL-6, and TNF- $\alpha$  have all been associated with CVD, systemic inflammation, and mental health problems such as depression (Bautista, Vera, Arenas, & Gamarra, 2005; Golia et al., 2014; Valkanova, Ebmeier, & Allan, 2013). There are many other pro-inflammatory cytockines as well as anti-inflammatory cytokines that can be measured in blood, with the caveat that a given cytokine may act as pro-inflammatory or anti-inflammatory under different conditions (Cavaillon, 2001). Less invasive methods of collecting inflammatory markers in blood are available, including the dried blood spot (DBS) technique (McDade, Williams, & Snodgrass, 2007). DBS can be used to measure a number of analytes, including CRP, IL-6, and TNF- $\alpha$  mentioned above (Massaro et al., 2019; McDade et al., 2007). Associations between childhood trauma and greater adult inflammatory markers CRP, IL-6, and TNF- $\alpha$  are reliably reported (Baumeister et al., 2016). However, effects of childhood adversity on inflammation in infancy through adolescence appear to be smaller. There appears to be enough developmental variability to detect small effects of childhood adversity on CRP and IL-6 in infants through adolescents (Kuhlman et al., 2019), though more studies with larger samples are needed. There is some evidence for heightened TNF- $\alpha$  levels in children who have experienced greater adversity (Dixon, Meng, Goldberg, Schneiderman, & Delamater, 2009; Wright et al., 2004), but there are very few studies that include TNF- $\alpha$ , and more studies are needed (Kuhlman et al., 2019).

Before using inflammation data, recent or current illnesses and infections must be probed with the child/adolescent and/or parent to rule out elevations in inflammatory proteins due to an illness or infection that could lead to incorrect conclusions. For CRP, values above 10 mg/L generally indicate an infection and should be removed. In adults, CRP levels below 1 mg/L indicate low cardiovascular risk, between 1 and 3 mg/L indicate intermediate risk, and above 3mg/L indicates high risk for future cardiovascular events (Pearson et al., 2003). There are no similar adult cutoffs for IL-6 and TNF- $\alpha$ . For children and adolescents, there is currently no consensus on what level of CRP, IL-6, or TNF- $\alpha$  confers risk for later CVD. As a result, CRP, IL-6, and TNF- $\alpha$  are often analyzed continuously in studies with children and adolescents. It is also important to note that inflammation is generally higher in individuals with higher levels of fat mass (Park, Park, & Yu, 2005), so it is important to consider both fat mass and inflammation in conceptualization and analysis.

#### 5.5 | Metabolic syndrome (MetS)

In adulthood, MetS is defined as having at least three of the following five factors: high waist circumference, high triglycerides, low HDL, high blood pressure, and high fasting glucose. Adult MetS is a strong predictor of future CVD and diabetes (Ford, 2005). In children and adolescents, MetS is difficult to define, and the clinical relevance is unclear as few longitudinal studies have followed children into adulthood to assess whether MetS in childhood predicts clinical outcomes in adulthood (Magge, Goodman, & Armstrong, 2017; Steinberger et al., 2009). There is evidence in children and adolescents of clustering in adverse levels of MetS factors and in longterm rates of change into adulthood (Chen, Srinivasan, Li, Xu, & Berenson, 2007). In fact, over 40 different definitions for pediatric MetS have been used in the literature, and there is currently no consensus on the definition among groups such as the WHO, National Heart, Lung, and Blood Institute, and the International Diabetes Foundation (Al-Hamad & Raman, 2017). Rather, it may be more important to identify clustering of individual cardiovascular risk factors, such as high blood pressure or high lipid levels, in children and adolescents rather than categorical classification of with or without MetS (Magge et al., 2017). It may also be more helpful to examine components of MetS on a continuum in children rather than use cutpoints for individual factors. The AAP recommends screening for and treating individual risk factor components of MetS in children.

Recently, researchers collaborating with the AHA developed guidelines for ideal cardiovascular health in children and adolescents, which includes both biological (e.g., BMI percentile, fasting glucose) and behavioral measures (e.g., dietary quality, physical activity) (Lloyd-Jones et al., 2010). Framing cardiovascular health in a more positive way and aiming to promote health rather than mitigate risk may be especially helpful in pediatric populations where CVD is rare. The field of pediatric preventive cardiology focuses on reducing these risk factors to improve pediatric cardiovascular health. Pediatric preventive cardiologists are often found at large medical centers and may be good collaborators for conducting collaborative research along with SEED scientists.

#### 5.6 | Allostatic load

To this point, we have been utilizing the DOHaD framework to guide our background and methods review. However, we also want to address the allostatic load model as a model that makes predictions about an organism's adaptation to various levels of stress (McEwen, 1998). The allostatic load model predicts that with repeated exposure to severe stress, the body begins to show "wear and tear" that leads to poorer mental and physical health outcomes (McEwen, 1998). This process of wear and tear occurs through the body's normal process of allostatis, which is the body's process of achieving stability while adapting to challenges (McEwen, 1998). Mediators of allostasis include the ANS, HPA axis, cardiovascular, immune, and metabolic systems (McEwen, 1998). Researchers have developed ways to assess the wear and tear, or allostatic load, on an individual through measuring several aspects of physiology. Measuring allostatic load rather than a specific outcome such as cardiovascular risk allows researchers to measure underlying physiological risk, which is hypothesized to increase risk for an array of mental and physical health problems (Juster, McEwen, & Lupien, 2010). As children and adolescents are less likely than adults to have diseases associated with aging, using allostatic load may be especially appealing in children and adolescents to measure underlying risk for later disease. There is currently no gold standard method of measuring allostatic load in children or adolescents. However, it is generally recommended that biomarkers from each of the cardiovascular, metabolic, and immune systems be included (Duong, Bingham, Aldana, Chung, & Sumner, 2017). Typically, there are multiple individual biomarkers that are used to create a continuous composite score of allostatic load by summing the number of individual biomarkers that are classified as being at a level of risk based on established guidelines or being high risk within the sample (Calcaterra et al., 2019; Duong et al., 2017; Theall, Drury, & Shirtcliff, 2012). Individual biomarkers in children

and adolescents have included: HDL-C, LDL-C, cholesterol, triglycerides, fasting glucose, insulin resistance, waist circumference, BMI percentile or *z*-score, CRP, glycosylated hemoglobin (measure of blood sugar control, which is important for diabetes management), blood pressure, and asthma diagnosis (Calcaterra et al., 2019; Rainisch & Upchurch, 2013; Theall et al., 2012).

#### 5.7 | Assessing cardiometabolic risk in infancy

Although much work has tracked the associations of stressful life events and environmental adversity in childhood to chronic disease outcomes that emerge much later in life, it may be possible to detect cardiometabolic risk signatures early in life, decades before the onset of overt disease. Low birth weight and catch-up growth in infancy have been identified as independent risk factors for obesity in adolescence and later adulthood (Ong, 2006). Obesity is both difficult to reverse once established and is on the causal pathway to cardiometabolic disease. Thus, assessment of adiposity and rapid weight gain are among the most relevant aspects of cardiometabolic risk during infancy. Low birth weight and earlier age at adiposity rebound are also associated with increased risk of type 2 diabetes and hypertension later in life (Mi, Fang, Zhao, & Zhong, 2017; Rolland-Cachera et al., 1984).

Acute stress has a strong effect on the inflammatory response, which can be detected by measuring circulating pro-inflammatory cytokines such as IL-6, interleukin-1B, TNF- $\alpha$ , or CRP (Marsland, Walsh, Lockwood, & John-Henderson, 2017). A few studies (mostly in resource-poor settings) have shown that elevated levels of these pro-inflammatory markers in infancy predict growth faltering (Syed et al., 2018), while at least 25 years of research supports chronic subclinical inflammation as a central player in the causal pathway of atherosclerosis, diabetes, cognitive decline, and other conditions of aging (Tracy, 2003). The key role of inflammation in stress, growth restriction, and chronic disease suggests that measurement of inflammatory markers such as IL-6 and CRP in infants could be useful early biomarkers to include in studies tracing the health implications of early psychosocial stress. However, more research on levels of these inflammatory biomarkers in infancy while the immune system is developing in relation to psychosocial factors and later cardiovascular health are needed.

### 6 | CONCLUSIONS

The incorporation of anthropometry and measures of cardiometabolic health into SEED studies offers exciting avenues of transdisciplinary research. We hope that this review provides researchers with ideas for how to incorporate these measures into SEED research and supplies information about important considerations, resources, and collaborators for adding these measures. We acknowledge that this review is by no means comprehensive, and we have attempted to direct readers to appropriate review papers whenever possible. We include a link to resources we have compiled on the Open Science Framework (https://osf.io/4yrf2/). In this review, we have covered some of the gold standard measures but also want to emphasize that the feasibility of performing these measures depends on the goals of the study. High-risk populations or resource-poor settings may make gold standard methods impractical. However, there are still options for low-cost and feasible methods to incorporate some of these measures into research (see Figure 1), such as height/length, weight, and head and waist circumference. As discussed, these measures can provide valuable insight into nutrition status (e.g., under- or overnutrition) and cardiometabolic risk. We also want to emphasize the importance of assessing inter-rater reliability and receiving ongoing training in all of the methods to assess cardiometabolic risk discussed above in order to obtain accurate data (De Miguel-Etayo et al., 2014).

Each of the authors has had the opportunity to work across traditional disciplinary boundaries, and we believe that transdisciplinary work is needed to address some of the biggest challenges in SEED science. Children who experience greater psychosocial stress are more likely to struggle with malnutrition, obesity, and CVD compared to children who experience less psychosocial stress, which makes these issues especially important for SEED researchers. We also encourage SEED researchers to pose their research questions in the context of DOHaD theory, which may inspire additional research understanding how factors may be embedded across development to influence adult health and disease. Promising future research directions include: (a) identifying levels of cardiometabolic risk markers in childhood and adolescence that reliably predict adult CVD; (b) better understanding how nutrition and diet, nutrition status, and psychosocial stress jointly affect cognition, emotion, and health; (c) understanding the neurobiological, cognitive, behavioral, and socioemotional predictors of health in the context of childhood stress; (d) assessing how trajectories of child development and health interact over time; and (e) designing and testing behavioral interventions to reduce child obesity (Lumeng, Taveras, Birch, & Yanovski, 2015) and improve cardiovascular health. We hope this review serves as a resource for researchers who wish to add measures of growth and cardiometabolic risk to their research or who want to better understand these measures when reading transdisciplinary research.

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#### CONFLICT OF INTEREST

The authors have no conflicts of interest to report.

#### DATA AVAILABILITY STATEMENT

There are no associated data for this review.

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O'BROKTA ET AL.

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