The Brain and Early Experience Study: Protocol for a Prospective Observational Study

William Roger Mills-Koonce¹, BA, PhD; Michael T Willoughby², BSc, PhD; Sarah J Short³, BSc, PhD; Cathi B Propper⁴, BSc, PhD

¹School of Education, The University of North Carolina at Chapel Hill, Chapel Hill, NC, United States
²Research Triangle Institute, Research Triangle Park, NC, United States
³School of Education, University of Wisconsin at Madison, Madison, WI, United States
⁴Frank Porter Graham Child Development Institute, The University of North Carolina at Chapel Hill, Chapel Hill, NC, United States

Abstract

Background: Children raised in conditions of poverty (or near poverty) are at risk for nonoptimal mental health, educational, and occupational outcomes, many of which may be precipitated by individual differences in executive function (EF) skills that first emerge in early childhood.

Objective: The Brain and Early Experience study considers prenatal and postnatal experiences that may mediate the association between poverty and EF skills, including neural substrates. This paper described the study rationale and aims; research design issues, including sample size determination, the recruitment strategy, and participant characteristics; and a summary of developmental assessment points, procedures, and measures used to test the study hypotheses.

Methods: This is a prospective longitudinal study examining multiple pathways by which poverty influences normative variations in EF skills in early childhood. It is funded by the National Institute of Child Health and Human Development and approved by the institutional review board.

Results: Recruitment is complete with a sample of 203 participants, and data collection is expected to continue from September 2018 to February 2024. Of those recruited as low socioeconomic status (SES), 71% (55/78) reported income-to-needs (ITN) ratios of <2.0, and 35% (27/78) reported ITN ratios of <1.0. Among participants recruited into the not-low SES stratum, only 8.8% (11/125) reported ITN ratios of <2.0, and no participant reported ITN ratios of <1.0. The average ITN ratio for participants recruited into the low-income stratum was significantly lower than the average for the high-income recruitment cell (P<.001). Comparable recruitment outcomes were observed for both Black and non-Black families. Overall, the sample has adequate diversity for testing proposed hypotheses, with 13.3% (27/203) of participants reporting ITN ratios of <1 and >32.5% (66/203) reporting ratios of <2.0.

Conclusions: Preliminary results indicate that the recruitment strategy for maximizing variation in family SES was successful, including variation within race. The findings of this study will help elucidate the complex interplay between prenatal and postnatal risk factors affecting critical neurocognitive developmental outcomes in early childhood.

International Registered Report Identifier (IRRID): DERR1-10.2196/34854

(JMIR Res Protoc 2022;11(6):e34854) doi: 10.2196/34854

Keywords

poverty; executive functioning; parenting; language; sleep; neurological development
Introduction

Background

On average, children raised in poverty perform more poorly on cognitive assessments and achieve lower academic outcomes than children from higher-income families [1], possibly owing to the effects of nonoptimal environmental experiences compromising early brain maturation and executive function (EF) skills [2]. Despite its significance, limited information exists on the neural and cognitive precursors and the social determinants of EF skills, especially during the first 3 years of life [3]. To address this scientific gap, the Brain and Early Experience (BEE) study examines specific prenatal and postnatal pathways from economic disparity to EF skills mediated by environmental experience and early structural and functional brain development.

Study Design Considerations

Beginning in utero, poverty imposes numerous risk factors [4] that contribute to an achievement gap persistent throughout formal schooling for poor and nonpoor children [5]. This leads to less educational attainment, increased likelihood of single parenthood, lower occupational status, poorer physical and mental health, and increased risk for all causes of mortality [6]. The limited development of neural substrates supporting neurocognitive development and the disrupted or delayed emergence of EF skills and other cognitive and language abilities may serve as mechanisms by which poverty gets under the skin and alters developmental trajectories across the life span [7].

A variety of cognitive processes that support goal-directed behavior are subsumed under the construct of EF. In early childhood, the 3 most widely studied subdomains of EF include working memory, defined as the holding in mind and updating of information while performing some operation on it; inhibitory control, defined as the inhibition of prepotent or automatized responding when engaged in task completion; and mental flexibility, defined as the ability to shift attentional or cognitive set among distinct but related dimensions or aspects of a given task [8]. Identifying the specific experiential and neurocognitive mechanisms through which poverty leads to poor EF is essential for optimizing early intervention programs.

Neuroscientists investigating how poverty influences children’s neural development have identified systematic differences in structural brain development that mediate associations between poverty and impaired academic outcomes [9-12]. Specific examples include developmental differences in the maturation of frontal and temporal lobe gray matter that explained up to 20% of the variance in low-income children’s cognitive deficits [13] and different surface-based morphometry indexes (ie, cortical thickness, surface area, cortical folding, and combinations of these) between poor and not-poor children [3]. These brain regions (and associated neural networks) may be highly vulnerable to the early environmental risks associated with poverty. Therefore, the BEE study applies a developmental science approach to the study of prenatal [14] and postnatal [15] determinants of neurocognitive development and later EF.

As the earliest experience in development, the prenatal period is a highly sensitive time for neurocognitive development. Although there are numerous studies on women’s physical health during pregnancy and offspring neurodevelopment [16-18], there is less research on how mental health and psychological experiences during pregnancy may be associated with subsequent neurodevelopmental outcomes. Factors such as elevated prenatal stress may influence early neurocognitive skills through their influence on the proliferation, differentiation, migration, and aggregation of fetal neurons [19,20]. For example, Buss et al [21] report that pregnancy-specific anxiety predicts reduced gray matter density in the cortex, the left middle temporal lobe, the entorhinal cortex, and the parahippocampal gyrus at 6 to 9 years of age. The BEE study examines both subjective measures of prenatal stress (via self-report) and objective measures (via biological stress markers) as potential mediators of associations between poverty and prenatal brain development.

Following birth, poverty adversely influences children’s development through early proximal experiences [22], with negative associations reported between family socioeconomic status (SES) and caregiving behaviors [23,24], child sleep quality [25], and child language exposure [26,27]. In contrast, multiple studies indicate that the quality of early caregiver–child interactions predict childhood EF and changes in EF throughout time [28-30]. These effects may be owing to the support and stimulation provided by a responsive parent or the favorable environment in which children can practice these developing skills as active agents in their own learning and skill acquisition. Similarly, sleep hygiene in early childhood predicts emerging EF in children [31]. Bernier et al [32] report that sleep in the first year of life predicts improved EF at 26 months; however, it was not related to other cognitive outcomes such as verbal ability or broader cognitive functioning, suggesting that sleep may be particularly important for EF. Activities that engage executive control are effortful and may be supported through energy restoration that occurs during sleep [33,34]. Finally, early language exposure contributes to children’s emerging language and EF skills [35,36]. Children’s expressive language contributes to problem solving and self-directed speech to regulate thoughts, emotions, and behavior [37,38] and emerging EF skills [39-44].

Considering these findings, the BEE study examines caregiver–child interaction quality, child sleep hygiene, and language exposure as environmental experiences potentially mediating the associations between poverty, brain development, and emerging EF. These constructs were also selected because they (1) are among the most salient for all children and account for a large percentage of the child’s daily lived experiences, (2) have reliable, valid, and developmentally appropriate methods of measurement across time, and (3) are moderately stable throughout time, providing confidence that our assessments will represent the overarching experience of the first 3 years of life. It is also important to note that extreme levels of poverty are not necessary for observing the associations between family income and child outcomes. Several studies report that children living above but near the poverty level—or even up to 2 times the poverty level—experience significant neurocognitive risk
As described further, the BEE study purposefully sampled families along a range of socioeconomic risks (with oversampling for those in poverty) to capture the variability in poverty and poverty-related risk predictive of neurological and EF development.

The BEE study also uses state-of-the-art functional magnetic resonance imaging (fMRI) measurements to observe neurological development soon after birth and before the third year of life. Most studies examining associations between poverty and the brain used global metrics of brain development (e.g., total gray matter and total surface area) and relied on cross-sectional data and mixed age samples. However, the development of EF skills involves efficient information processing between brain regions relying on the integrity of specific white matter tracts and the underlying neural networks connecting them [45]. Moreover, it may be that the developmental pattern during this earliest period of life is most informative, given that the most prolific changes in brain development that inform early EF occur from birth to 3 years [46]. The BEE study addresses these limitations by examining associations between poverty and early structural (i.e., white matter integrity via diffusion tensor imaging [DTI]) and functional (i.e., resting state [rs] networks via fMRI) aspects of brain development at 2 weeks and 30 months of age. In this study, we will interpret brain development at 2 weeks of age as a product of prenatal experiences and brain development at 30 months of age as a combination of prenatal and postnatal experiences.

Aims and Hypotheses of the BEE Study

The BEE study addresses 3 specific aims. Aim 1 involves the examination of associations between poverty and neonatal structural and functional brain development at 2 weeks of age. Two sets of hypotheses are proposed for aim 1. The first is that poverty will predict individual differences in neonatal structural brain development (specifically for white matter tracts that support cognitive processes of emerging EF) and functional brain development (including rs networks related to attention, salience, executive control, and default mode); the second is that these associations will be partially mediated by prenatal experiences, including stress, nutrition, obesity, and toxic environmental exposures. Aim 2 involves the examination of associations between poverty and toddler structural and functional brain development at 30 months of age and potential mediators of these associations. Three sets of hypotheses are proposed for aim 2. The first is that poverty will predict individual differences in changes in toddler structural and functional brain development. The second hypothesis is that prenatal experiences will partially mediate these associations. The third hypothesis is that postnatal experiences (i.e., caregiving, sleep hygiene, and language exposure) will partially mediate the association between poverty and toddler brain development. Finally, aim 3 involves the examination of pathways from poverty to EF at 36 months of age. Three sets of hypotheses are proposed for aim 3. The first hypothesis is that poverty will be negatively associated with EF, the second is that prenatal experiences will partially mediate the association between poverty and EF, and the third is that postnatal experiences will also partially mediate these associations. The path model in Figure 1 illustrates these aims and hypotheses.

Methods

Sample

Power Analyses to Determine Sample Size

Two Monte Carlo studies were conducted to determine the statistical power to evaluate study aims given the proposed sample size and design. Both studies, which adopted the approach of Thoemmes et al [47], included a population generating model of 200 participants, assumed a type I error rate of 0.05, and involved 5000 replications. With these parameters, for tests of main effects (encompassing associations between poverty child outcomes, including brain development at 2 weeks and 30 months and child EF, general cognitive ability, and language development at 30-36 months), the sample has the power (≥0.98) to detect small to medium (R²=0.075) sized main effects but has limited power (0.51) to detect small (R²=0.02) main effects. For tests of mediation (encompassing
the main effects as mediated by prenatal and postnatal experiences), the sample has power (>0.99) to detect single mediated effects when both the \( a \) and \( b \) paths are of medium size (\( R^2 = 0.13 \)). The study has power (>0.89) to detect a mediated effect when both \( a \) and \( b \) paths are of small to medium size (\( R^2 = 0.075 \)). However, the power to detect a mediated effect drops to 0.45 when either \( a \) or \( b \) paths are of small size (\( R^2 = 0.02 \)), even if the other path is of medium size. As a point of reference for those readers who are unfamiliar with Monte Carlo methods, 200 results in a power of 0.80 to detect bivariate correlations [\( r \] ≥ 0.20, which also corresponds to a small to medium-sized effect per the Cohen conventions [48]. The study will rely on high-quality measurement and repeated measures to improve the magnitude of the effects.

Recruitment Strategy

From August 1, 2018, through October 31, 2020 (26 months), we used multiple methods to reach the population of interest (pregnant women in their second trimester). These include targeted advertisements on social media, online community message boards, hospital records in the local university-based medical center, listserv emails to local university staff and students, paper fliers posted in obstetrician–gynecologist offices in the local geographic area, and personally staffing information tables at the Women, Infant, and Children centers before, during, and after local pregnancy and birthing classes. Individuals interested in participating in the study were directed to a brief web-based survey that asked questions targeting inclusion criteria, including being currently pregnant with a single fetus and speaking primarily English at home. If both criteria were met, subsequent questions regarding due date, contact information, and permission to be contacted for participation were asked. Next, a study personnel used this information to schedule a phone call with each participant. During this phone call, the researcher confirmed the inclusion and exclusion criteria and obtained the person’s address to confirm that they lived within a 45-minute radius of the study location and that there was no intent to move out of the geographic area in the next 3 years.

The research staff then asked demographic questions to assign potential participants to 1 of 4 recruitment cells based on a 2×2 (SES×race) design. This categorization as part of the recruitment process was important because, as a study focusing on the role of poverty in early development, we sought to limit confounding between income and race by oversampling (relative to local family demography) for combinations of (1) lower SES and non-Black racial identity and (2) not-low SES and Black racial identity. Three demographic questions were asked for all potential participants who met inclusion criteria. The first question was about the person’s race and ethnicity. The second question was about the individual’s highest level of education. The third question asked about the use of a range of federal social services that are eligible to families at or below 185% of the federal poverty level, including Women, Infant, and Children program, Temporary Assistance for Needy Families, Supplemental Nutrition Assistance Program, or Medicaid. Following the precedent established by other large-scale developmental studies of families living in poverty [49], we used these questions to assign potential participants into a low-SES group if they had no education beyond a high-school diploma or general education degree or if they currently used a social service. Otherwise, they were classified as not-low SES. We designated a participant as Black if they self-identified as Black, African American, or multiracial Black; otherwise, they were designated as non-Black. These 2 dimensions were cross-tabulated to create 4 target recruitment cells with the goal of recruiting comparable numbers of low-SES and high-SES families within each racial group to reduce potential confounding between race and family income. The assignment of families into cells was intended solely to guide recruitment efforts and should not be used for hypothesis testing regarding family income and race. Families were considered formally enrolled if they completed 2 of the first 3 data collection visits (including the prenatal visit, the 2-week postnatal visit, and the 6-month postnatal visit). If a family did not meet these inclusion criteria, they were dropped from the study and replaced by a newly recruited family with comparable SES and racial identities. See Multimedia Appendix 1 for a flowchart documenting recruiting and sampling numbers that resulted in the final sample size of 203 participants.

Sample

On average, biological mothers of BEE participants were aged 30.53 (SD 5.35, range 18-46) years and had completed 15.36 (SD 2.66) years of education at the prenatal visit. Approximately 33% (67/203) of the sample identified as Black or African American, and approximately 8.9% (18/203) identified as Hispanic or Latina. At the prenatal visit, 90.1% (183/203) of mothers were currently in a romantic relationship, 66.9% (136/203) were married to their partner, and 84.06% (170/202,223) were living with their partner, defined as spending a minimum of 3 nights together at the same residence.

Design and Procedures

Overview

The COVID-19 pandemic had 3 impacts on the design and execution of the BEE study. First, the pandemic interrupted and prolonged study recruitment procedures resulting in a recruitment window of 26 months. Second, some of the in-person planned visits were adapted to be conducted on the web and involve family-led data collection activities. This was most disruptive during the prenatal, 2-week, and 6-month visits that had already begun as in-person protocols and were shifted to web-based protocols because of the COVID-19 pandemic. Third, the timings of some visits were delayed. Although the original plan included 15- and 24-month visits, the final data collection timeline shifted these visits to 18 and 30 months. Each of these issues is discussed in the context of a brief synopsis of the overall study timeline.

Following recruitment, pregnant women are seen at a prenatal laboratory-based visit. Research staff follow up with participants around the expected due dates to speak with the participant as soon as possible following the child’s birth. During this conversation, staff determines if any new exclusion criteria for participation in the study are present—this includes being born with low birth weight (<2500 grams) or a gestational age below...
Pregnant women were invited to participate in their first data collection visit for the BEE study at the Biobehavioral Laboratory at the University of North Carolina (UNC) at Chapel Hill School of Nursing. Following a description of the overall study and the prenatal data collection protocol, informed consent for participation was obtained. Next, participants were interviewed by research staff, completed a series of questionnaires, a series of computer-based cognitive assessment tasks, and engaged in a qualitative interview about their experiences during pregnancy and expectations about motherhood. Finally, the research staff collected hair, blood, saliva, and urine samples that were immediately processed and placed in cold storage at −80 °C. Participants were compensated for their travel costs and provided US $50 for their time. For this visit protocol, some visits were conducted in person in the laboratory, and others were done via Zoom call because of the human participants contact restrictions related to the COVID-19 protocols. The same tasks and instructions are used in each visit, but the parent–child interaction tasks were performed in person in the laboratory, and participants returned the materials to the study via mail.

**The 6-Month Home Visit (and Web-Based Visit During the COVID-19 Pandemic)**

Participants are next seen during a home visit at 6 months of age (some were seen only via video-based visits on the web because of the COVID-19 pandemic) and then on a video-based visit on the web at 18 months of age (because of the COVID-19 pandemic). Next, participants are seen for a video-based visit on the web at 30 months of age, followed immediately by a laboratory visit also at 30 months of age (for the second fMRI scan). Finally, participants are seen at the laboratory for their last data collection visit at 36 months of age.

**Prenatal Laboratory Visit (and Web-Based Visit During the COVID-19 Pandemic)**

During the COVID-19 Pandemic) The 2-Week Laboratory Visit (and Web-Based Visit

Participants are next seen during a home visit at 6 months of age (some were seen only via video-based visits on the web because of the COVID-19 pandemic) and then on a video-based visit on the web at 18 months of age (because of the COVID-19 pandemic). Next, participants are seen for a video-based visit on the web at 30 months of age, followed immediately by a laboratory visit also at 30 months of age (for the second fMRI scan). Finally, participants are seen at the laboratory for their last data collection visit at 36 months of age.

**The 6-Month Home Visit (and Web-Based Visit During the COVID-19 Pandemic)**

Research staff visit participants in their homes for the 6-month home visit. After providing informed consent for the visit, mothers answered interview items and completed a series of questionnaires, and then the mother and child participated in 2 parent–child interaction tasks. The first involves playing together with a standardized set of toys for 10 minutes; the second involves the mother going through a wordless picture book with her child for 5 minutes. Both tasks are video and audio recorded for later behavioral coding. After these tasks, the research staff explain the extended data collection protocols for child sleep hygiene (7 days) and language exposure (2 days) after the visit. To collect sleep hygiene data, mothers are provided with a wearable actigraphy device (Actiwatch-2) for the child and instructed on how to attach it to the infant’s ankle. Mothers are asked to leave the device on the child’s ankle for 7 full days, remove it only during bath times, and then reattach it once the child is dry. Mothers are also provided a sleep diary to complete every morning for the next 7 days. To collect language exposure data, mothers were provided with a Language Environment Analysis (LENA) recording device, 2 vests, and 2 daily diaries. Mothers are asked to identify 2 typical days (out of the next 7) in which they will be spending most of their time with their child. On those days, mothers are asked to dress their child as they normally would first thing in the morning, then turn on the LENA recorder and place it in the pocket of the vest before putting the vest on the child. Mothers are asked to leave the device on all day. These steps are repeated for another day during the 7-day period. Mothers are also asked to complete daily diaries about the contexts and people in the child’s environment during those days. Both the actigraphy and the LENA devices are retrieved from the participant’s home on the seventh day following the 6-month home visit. Participants were compensated for their travel costs and provided US $50 for their time.

For this visit protocol, some visits were conducted in person in the participant’s home, and others were done via Zoom call because of the human participants contact restrictions related to the COVID-19 protocols. The same tasks and instructions are used in each visit, but the parent–child interaction tasks were recorded using Zoom instead of in-person cameras. For participants who needed internet or computing devices for the Zoom call, we provide tablets and wireless hot spots to facilitate the web-based visit. For those receiving the web-based protocol, wearable devices and biospecimen collection materials were

https://www.researchprotocols.org/2022/6/e34854
mailed to the home, and participants returned the materials to the study via mail.

**The 18-Month Remote Visit**

The research staff invited participants to join a video call for the 18-month Zoom visit. After providing informed consent (electronically) for the visit, mothers answer interview items and complete a series of web-based questionnaires. Wearable devices and biospecimen collection materials were mailed to the home, and participants returned the materials to the study via mail. Participants were compensated US $50 for their time.

**The 30-Month Remote Visit**

The research staff invited participants to join a video call for the 30-month Zoom visit. After providing informed consent (electronically) for the visit, mothers complete a series of questionnaires, and then mother and child participate in a parent–child interaction task. This task involves presenting the child with a series of 3 peg puzzles of increasing difficulty and instructing the mother that she can provide any assistance that she chooses. The task is video and audio recorded for later behavioral coding. The research staff explain the extended data collection protocols for child sleep hygiene (7 days) and language exposure (2 days) that continue after the visit. Additional components of this study visit are identical to those protocols used in the 6-month home visit described earlier (eg, actigraphy, LENA, and biomarkers). Wearable devices and biospecimen collection materials were mailed to the home, and participants returned the materials to the study via mail. Participants were provided US $50 for their time.

**The 30-Month Laboratory Visit**

Participant mothers and children came to the UNC Biomedical Research Imaging Center for the 30-month laboratory visit for their second fMRI scan. As at the 2-week visit, this scan occurs while children are naturally sleeping. To facilitate this, the 30-month visits are scheduled during the evening at the child’s usual bedtime. After providing informed consent for the visit, mothers complete a short series of questionnaires, and children’s natural bedtime routine is replicated to facilitate child sleep. Next, the research staff administer the Receptive Vocabulary, Information, Block Design, and Object Assembly subscales of the Wechsler Preschool and Primary Scale of Intelligence (the former 2 subscales index vocabulary acquisition and the latter 2 index visual-spatial abilities) [50]. Mothers apply ear protection when they determine it will be most tolerable for their child. At this point, the research staff follow the same fMRI protocol as used during the 2-week laboratory visit. Participants were compensated for their travel costs and provided US $125 for their time.

**The 36-Month Laboratory Visit**

Participant mothers and children came to the Biobehavioral Laboratory at the Frank Porter Graham Child Development Center at UNC Chapel Hill for the 36-month laboratory visit. After providing informed consent for the visit, mothers answer interview items and complete a series of questionnaires. During this time, research staff assess children’s EF using the EF Touch battery [51] and assess children’s language using the Peabody Picture Vocabulary Test [52]. Participants were compensated for their travel costs and provided US $50 for their time.

**Measures**

A list of all interviews and questionnaires administered at each data collection visit is provided in Multimedia Appendix 2. In the further sections, we describe the measurement of each key construct used to address the specific aims 1, 2, and 3 in greater detail.

**Household Income**

At each data collection visit, mothers reported (1) their annual income, (2) the annual income of their partner (if co-residing in the home), (3) the annualized contributions to the household of all others in the household, and (4) the annualized contributions from other sources of income (eg, unemployment insurance, worker’s compensation, and social security retirement). Using this information, an annual household total income variable is created by summing all sources of income. Next, we divide this total amount by the federal poverty threshold for a family of that particular size and composition to create the income-to-needs (ITN) ratio—a standard measure of a family’s economic situation where a value of 1.0 indicates living right at the poverty line. This approach to quantifying household income has been used extensively in population-based studies of poverty and child development [49].

**Prenatal Stress**

**Self-report of Subjective Stress Experience**

In all, 3 self-report questionnaires on experiences of stress are completed at the prenatal visit. The Cohen Perceived Stress Scale is a 10-item questionnaire measuring general perceived stress during the past month [53]. The items are rated on a 5-point Likert scale ranging from never (0) to very often (4) and have demonstrated high reliability; the reliability of Perceived Stress Scale in this study is 0.88. The Pregnancy-Related Anxiety Questionnaire–Revised-2 is a 10-item measure assessing pregnancy-related stress [54]. The items are rated on a 5-point Likert scale ranging from 0=definitely not a concern at all to 4=definitely a very big concern. The reliability of this scale in this study is 0.85. A modified 6-item version of the Economic Strain Questionnaire measures financial stress, including concerns about the inability of families to make ends meet and not having enough money for a home, clothing, food, and medical care [55]. Items are rated on a 5-point Likert scale (0=great deal of difficulty to 4=none difficulty at all) and on a 4-point Likert scale (0=strongly disagree to 3=strongly agree). Previous studies report adequate reliability of this scale and suggested that all 6 items could be standardized and summed or averaged to create a global measure of economic strain [55,56]. The reliability of this scale in this study is 0.88.

**Biological Indexes of Prenatal Stress**

A total of 2 classes of biological measures of stress were collected during the prenatal visit. The first includes hair cortisol concentration measured from hair strands close to the scalp from the posterior vertex area of the participant’s head (this area has shown the lowest coefficient of variation [CV]). The 3 cm of hair closest to the hair roots are analyzed [57], reflecting...
exposure during the last 3 months (based on the hair growth rate of approximately 1 centimeter per month) [58]. Cortisol is measured in methanol extracts of hair using a competitive radioimmunoassay following standard assay procedures. The intra-assay CV is 1.17%, and the interassay CV is 5.12% in this sample—both values indicate reliable assaying results.

The second class of biological stress indexes includes multiple inflammatory markers observed during the prenatal visit. A total of 2 EDTA tubes are used to conduct antecubital venipuncture to collect nonfasting plasma samples from participants. Plasma samples are assayed for cytokines using a Meso Scale Discovery multiplex kit; this panel includes interleukin-6, interleukin-10, tumor necrosis factor, interleukin-1RA, interleukin-2, and interleukin-8, each measured in pg/mL. C-reactive protein is assayed using Meso Scale Discovery single-plex kit and is measured in mg/L.

Early Postnatal Experience

Caregiving

Video recordings of parent–child interactions will be coded by trained and certified coders to rate caregiving behaviors on the following dimensions: sensitivity, intrusiveness, detachment, stimulation of cognitive development, positive regard, negative regard, and animation. Sensitivity indexes the degree to which the caregiver is attuned and responsive to the physical and emotional needs of the child. Intrusiveness indexes the degree to which the caregiver is controlling and imposes their own agenda on the activity of the child. Detachment indexes the degree to which the caregiver is emotionally and physically detached and uninvolved with the feelings and activities of the child. Stimulation of cognitive development indexes the degree to which the caregiver provides linguistic stimulation in a developmentally appropriate way and scaffolds the activity to maximize the child’s cognitive experiences of the task. Positive regard indexes the degree to which the caregiver directs feelings of warmth, love, and enjoyment toward the child. Negative regard indexes the degree to which the caregiver directly displays harshness and hostility toward the child. Animation indexes the level of energy and enthusiasm that the caregiver displays while interacting with the child. Each dimension is rated on a 7-point scale ranging from not at all characteristic of this caregiver to extremely characteristic of the caregiver. After being certified as reliable, each coder will continue to code a minimum of 20% of cases with a master coder for each coding assignment to prevent coder drift. Previous use of this parent–child interaction task and coding protocol has repeatedly identified 2 parenting composites guided by factor analyses [24,59], and we anticipate observing comparable factors. The first is referred to as sensitivity and is the mean of sensitivity, detachment (reversed), stimulation of cognitive development, positive regard, and animation. The second is harsh intrusiveness and is the mean of intrusiveness and negative regard.

Sleep Hygiene

A multi-method approach will be applied to assess sleep hygiene [60,61]. The actigraphy monitor worn on the child’s ankle contains an accelerometer that measures limb movement in 15-second epochs. At the end of the sleep assessment week, actigraphy data are downloaded to a computer and edited using Phillips Actiware software (version 6.0). The Actogram algorithm settings are as follows: immobile minutes for sleep onset were set to 5 minutes; minimum rest interval size was set to 20 minutes; multiple rest intervals per day were allowed; automatically set minor rest intervals were allowed. The activity threshold for scoring the child as awake is set to the automatic setting (0.888×average activity count)—both the algorithm and threshold for scoring sleep or wake state have been previously validated [62].

The output from the Actiware program includes a listing of all sleep and wake intervals. Infant sleep onset time is determined as the start time of the sleep interval closest to the caregiver-reported bedtime (see sleep diary description below). Similarly, the child’s rise time is determined as the end time of the sleep interval closest to the caregiver-reported rise time. Using sleep onset time and rise time, we subsequently calculate the duration of the child’s sleep period. Child sleep time in minutes is determined by summing infant sleep time in each sleep interval between sleep onset time and rise time. Child wake time in minutes is determined by summing infant wake time in each sleep interval during the sleep period. Child night wakings are determined by subtracting 1 from the number of sleep intervals during the nighttime sleep period (ie, if the child sleeps in 3 sleep intervals, there are 2 night wakings). Finally, the longest sleep period equals the duration of the longest sleep interval during the nighttime sleep period.

In addition to actigraphy measures, the parent completes daily sleep diaries for the child. Every day during the sleep assessment week, research staff call mothers to obtain information about the previous day’s naps and nighttime sleep, including number, location, and duration of naps, child bedtime, number of night wakings, types of interventions used during night wakings, and infant rise time [63]. Mothers also report any unusual occurrences that may have influenced the previous night’s sleep, such as child illness.

Language Exposure

The LENA digital recorder and software automatically processes the audio-based language environment the child experiences during the 2 days of data collection. As a wearable device, the LENA recorder collects recordings of the language environment for the entire day (16 hours; for the purposes of this study, wake time to bedtime). After 2 full days of data collection, the LENA recorder connects to a computer, and its software automatically uploads and analyzes the language data using a series of iterative modeling algorithms developed by the LENA Research Foundation. This process segments the recordings based on acoustic energy and generates 3 language measures mapping onto the Hart and Risley language exposure dimensions—adult word, conversational turn, and child vocalization counts [64]. Adult word count is the total number of adult words spoken near the child. Conversational turn count is the total number of conversational interactions the child engages in within an adult (this involves one person speaking and the other responding within 5 seconds). Child vocalization count is the total number of speech-like utterances produced by a child. LENA software
also generates other language indexes, such as overlapping speech, television and media, and background noise. This study focuses on adult word count and conversation turns as key indicators of child language exposure in the first 3 years of life.

**Brain Development**

DTI and rs fMRI scans are acquired at 2 weeks and 30 months of age. Scanning sequences are fully compatible with the Human Connectome Project (Multimedia Appendix 3). We use previously established imaging processing pipelines, quality control measures, and analysis protocols, which have proven successful for repeat scanning with infants and young children [65-68].

**DTI Analysis**

Diffusion images are screened using an automatic program for quantifying motion artifacts and corrupted sections using the diffusion-weighted imaging or DTI analysis quality control tool DTIPrep [69]. This program includes the correction of motion, eddy of current artifacts, and removal of outliers. Diffusivity property maps, such as fractional anisotropies, are estimated using standard weighted least square fitting [70]. The Brain Extraction Tool [71] is used for the skull stripping of all images. Unbiased atlas building [70] with large deformation diffeomorphic metric mapping registration [72] is used after a linear registration [73]. All data to be analyzed in this study are used to build the atlas. Transformations obtained from the atlas building are applied to warp the original tensor images to the atlas space, and the final DTI atlas is obtained by averaging the warped images. DTI tractography in atlas space yields all fiber bundles of interest [74].

Our established tractography pipelines generate quantitative DTI data for cohesive analysis of imaging data collected at 2 weeks and 30 months of age. Diffusion properties (fractional anisotropy, axial diffusivity, and radial diffusivity) are generated for white matter tracts hypothesized to support emerging EFs and language, including arcuate, uncinate, and anterior cingulum. Tractography algorithms with Runge-Kutta integration are performed in the atlas tensor image constructed from the unbiased DTI registration. The improved signal-to-noise ratio of participant-specific atlases (integrating data from 2 weeks to 30 months), deformed to a group atlas, allows reliable extraction of fiber bundles that would be hard to extract consistently from individual data (ie, arcuate). Fiber tracts are parameterized by length to represent diffusion properties as a function of location along the selected tracts [73,74].

**Resting State**

We will primarily use a seed-based approach to characterize the dynamic developmental changes of functional networks related to attention, salience, executive control, and default mode. Specifically, network-specific seeds will be defined according to our previous studies and used to generate functional connectivity maps of each network for the scans collected at 2 weeks and 30 months. Two measures of network integrity will be derived: mean functional connectivity and network maturation score. For the former, after defining the functional connectivity map of each network, individual clusters of significance will be extracted, and a cross-correlation matrix among all clusters within the same network will be calculated for each participant. The mean functional connectivity will be defined as the average of all pair-wise correlations within the network. This measure quantifies the individual functional network integrity in an age-adaptive fashion based on age-specific functional connectivity maps. For our measure of network maturation score, we will calculate a network maturation score using adult functional network topology as a reference to quantify the degree of maturation toward adult-like network topology [75]. On the basis of the same adult reference, this measure facilitates statistical comparisons across age groups and has been shown to be sensitive to SES during infancy [76]. Specifically, using the adult group-level significant functional networks as references, a binary mask was derived for each network. Subsequently, the within-network connectivity was defined as the mean functional connectivity strength within the mask, indicating the degree of within-network synchronization, whereas outside-network connectivity was the mean functional connectivity of areas outside the network mask, indicating the degree of outside-network specialization. Finally, based on a previously established network matching concept [77], the subtraction of the outside-network connectivity from within-network connectivity yields a network matching score, indicating the degree of similarity between the network in question and the adult reference network in terms of functional connectivity strength distribution of the whole brain. This network matching score will be used as an overall measure to quantify the maturation of individual networks.

To test the robustness of our results, a data-driven independent component analysis approach [78], which has been used extensively in previous studies, will be applied to extract the corresponding networks and subsequent network-level functional connectivity measures [79]. Identical analysis as listed above will be carried out based on independent component analysis–based measures to replicate our findings using a seed-based approach. Finally, given the controversy of the preprocessing step of global signal regression [80,81], our results will also be tested with or without this step to identify converging findings.

**EF Skills**

Children’s EF is measured at 36 months using the EF Touch, a computerized battery of EF tasks that have been iteratively developed over the last 10 years [82-84]. Each EF task takes 3 to 7 minutes to complete. A total of 2 warm-up tasks are typically administered first to acclimate children to using the touch screen. The reliability and validity of the EF Touch battery have been extensively documented, including with children aged as young as 3 years [85]. Mean accuracy across items within each task will index task performance.

Three tasks assess children’s inhibitory control: Spatial Conflict Arrows, Silly Sounds Stroop, and Animal Go/No-Go. Two tasks will assess children’s working memory at 36 months of age: Working Memory Span and Pick a Picture. One task will assess children’s attention shifting at 36 months of age: Something’s the Same. In addition to performance-based measures, assessors will complete the Preschool Self-Regulation Assessment
following the administration of tasks. The Preschool Self-Regulation Assessment consists of 28 items that are combined to form attentional control and positivity scales that reflect behavior during EF task completion [86].

Analysis Plan for Key Aims

Before conducting statistical analyses, the psychometric properties (ie, item to total correlations and \( \alpha \) coefficients) of all questionnaires will be evaluated, and the interrater reliability (ie, intraclass correlations and \( \alpha \) or \( \kappa \) coefficients) of measures that were based on observational coding will be documented. Descriptive statistics will be computed for all measures with an emphasis on distributions and outliers. Data transformations will be considered for variables appreciably skewed. When multiple measures are correlated within an assessment period (eg, caregiver-reported stressors during the prenatal visit), principal components and factor analyses will be used to create composite scores.

A structural equation modeling approach will be used to test all study aims. All structural equation modeling models will be estimated using a robust full information maximum likelihood estimator. The robust full information maximum likelihood estimator accommodates nonnormally distributed outcome variables and represents a statistical best practice for accommodating missing or unbalanced data [87,88]. To maximize power and avoid distributional assumptions, bootstrapped tests of indirect effects will be used to test questions of mediation related to aims 2 and 3 [89,90]. An exemplar path diagram corresponding to aims 1 to 3 is depicted in Figure 1 (residual variances or covariances are omitted to simplify the presentation).

Examination of direct and indirect effects will provide formal tests of aim 1. This will include testing a direct effect of prenatal poverty on brain development at 2 weeks of age and testing this path as an indirect effect mediated by subjective measures of prenatal stress (from self-report) and biological indexes of stress (indexed by hair cortisol and inflammatory markers).

Examination of direct and indirect effects will provide formal tests of aim 2. Direct effects from postnatal poverty (aggregated across 6, 18, and 30 months of age) on brain development at 30 months (controlling for comparable neurological indexes at 2 weeks of age) will test the association between poverty and brain development in the first 30 months of life. We will evaluate a measurement model for a latent caregiving environment variable. We will use multiple measures within each assessment period (eg, caregiver-reported stressors) to form a composite score of caregiver stress (DTI and rs metric, caregiving environment latent variable, prenatal stress measure, and prenatal and postnatal indicators of poverty (if prenatal and postnatal indicators of poverty are collinear, a single combined index will be used). The tests of direct and indirect effects of prenatal and postnatal poverty will provide a formal test of aim 3.

The previous analyses will provide definitive tests of all study aims. However, as described in the Measures section, we will also collect multiple indicators of general cognitive development (eg, Wechsler Preschool and Primary Scale of Intelligence and expressive language) at earlier assessments, and these interim measures will be used as outcomes in analyses that begin to test the guiding questions of this study before the completion of final data collection. These analyses will consist of simplified variations of the models described above. Interim measures will also be used for sample description.

Ethical Considerations

The study has institutional review board approval from the University of North Carolina at Chapel Hill (study number 17-1914).

Results

Recruitment is complete with a sample of 203 participants. Data collection began in September of 2018 and is expected to conclude by February 2024. This is a prospective longitudinal study; thus, analyses addressing the project’s specific aims are not yet available. However, analyses addressing the validity of the project’s recruitment strategy are provided by examination of the distribution of ITN ratios (a ratio of annual household income relative to a federal poverty threshold for a given household size, as described in the Measures section), the amount of money immediately available in savings or checking accounts, and homeownership data (all reported at the prenatal visit). We identified a participant as an outlier with an ITN ratio of 37.88; to limit the effects of this extreme value, we winsorized it to the next highest value of 15.79. The ITN distribution remained positively skewed with a minimum value of 0.00 and a maximum value of 15.79 (see Multimedia Appendix 4 for descriptive ITN information for the entire sample and separately by recruitment cell). For the total sample, 32.5% (66/203) of the participants reported an ITN ratio of <2.0 (interpreted as near poor or working poor), and 13.3% (27/203) of the participants reported an ITN ratio of <1 (interpreted as poor per the US federal definition of poverty). Next, we examined ITN distributions by recruitment SES designation. Of those recruited as low SES, 71% (55/78) reported ITN ratios of <2.0, and 35% (27/78) reported ITN ratios of <1.0. Among participants recruited into the not-low SES stratum, only 8.8% (11/125) reported ITN ratios of <2.0, and no participant reported ITN ratios of <1.0. The average ITN ratio for participants recruited into the low-income stratum was significantly lower than the average for the high-income recruitment cell (\( t_{188}=-9.643; P<.001; \text{Figure 2} \)).

Comparable percentages of income levels across SES designations at recruitment were observed for Black and non-Black participants. As seen in Figure 2, within racial categories, (1) non-Black low-income participants reported lower ITN ratios than non-Black high-income participants.
(\(P<.001\)), and (2) Black low-income participants reported lower ITN ratios than Black high-income participants (\(P=.002\)). Within SES categories, (1) non-Black low-income participants were not statistically different from Black low-income participants, and (2) non-Black high-income participants were not statistically different from Black high-income participants.

Although ITN is a broad indicator of a family’s financial standing, another indicator of economic insecurity is the availability of liquid funds, such as money in bank savings or checking accounts. According to the US Federal Reserve, in 2018, only 61% of Americans had immediate access to funds to cover a US $400 emergency expense [91]. In the study sample participating in this study, there are similar distributions and mean differences for participant reports of money immediately accessible in bank savings and checking accounts (Multimedia Appendix 5) compared with household ITN ratios. The average bank savings for the low-income recruitment cell was significantly lower than the average for the high-income recruitment cell (\(t_{181}=−5.44; P<.001\); Figure 3).

Although ITN and immediately accessible funds are important indicators of economic security, individual differences among families in overall wealth may also differentially buffer families from stress and negative life events (and racial disparities in wealth have been studied less than income despite being potentially larger in magnitude). An indicator of family wealth is homeownership. Among those recruited as high-SES, 76.8% (96/125) owned their homes compared with 20.8% (26/125) that rented their homes (note that these percentages do not sum to 100% because some participants lived with family and thus neither owned nor rented their homes). Conversely, among participants recruited as low-SES, only 8% (6/78) owned their homes, whereas 82% (64/78) rented their homes. Rates of homeownership for non-Black participants were similar to the overall sample. However, for Black participants, homeownership rates were lower. For high-SES Black families, only 35% (6/17) owned their own homes; for low-SES Black families, only 6% (3/48) owned their homes.

In summary, across these 3 indicators (ITN ratio, bank savings, and homeownership), there is convergent evidence validating the recruitment strategy’s goal of establishing a sample that is (1) economically diverse with adequate representation of low and very low-income participants and (2) economically diverse within Black and non-Black subsamples to reduce potential confounding between race and income.

**Figure 2.** Means and SEs (−2 to 2) for income-to-needs ratios across recruitment cells. (A) Socioeconomic status (SES)-only cells and (B) level and SES×race cells.
Figure 3. Means and SEs (−2 to 2) for bank savings across recruitment cells. (A) Socioeconomic status (SES)-only cells and (B) level and SES×race cells.

Discussion
The BEE study is one of the first prospective longitudinal studies of the prenatal and postnatal environmental influences on emerging EF in the first 3 years of life with specific foci on the potential effects of poverty, poverty-related stressors, and neurological substrates underlying cognitive abilities. Three primary aims examine (1) the associations between objective poverty and subjective prenatal experience on prenatal structural and functional brain development, (2) the associations between objective poverty and subjective postnatal experience on structural and functional brain development during the first 2.5 years of life, and (3) the associations between objective poverty and subjective postnatal experience on EF at 3 years of life. As described in this paper, several accommodations to this study were made in response to the COVID-19 pandemic, including prolonged participant recruitment, shifts to web-based data collection for some measurements, and alternation in the timing of visits and assessments. These accommodations have minimized the potential threats posed by the COVID-19 pandemic to the internal validity of the study. Despite the prolonged recruitment period, the results support the validity of the overall recruitment strategy, which was designed to establish a socioeconomically diverse sample with adequate variability in family incomes within both Black and non-Black families to reduce the potential confound of race and income. The web-based data collection of questionnaire data and observational protocols and the remote-based data collection of language exposure and child sleep hygiene have resulted in data comparable (qualitatively and quantitatively) with in-person data collection protocols. Finally, the changes in the timing of data collection visits maximized the number of participants that could participate in fMRI data collection while remaining within the targeted developmental window for examining neurological development pertinent to emerging EF within the first 3 years of life. We believe that the data being collected and subsequent analyses addressing the specific aims of this study will not be compromised by the COVID-19 pandemic.

Acknowledgments
The Brain and Early Experience study is funded by the National Institute of Child Health and Human Development (grant R01 HD091148). The authors would like to thank Marie Camerota, Ilana Berman, and Rebecca Stephens for their invaluable contributions as project directors and investigators on this study.

Authors’ Contributions
CBP and SJS are principal investigators for this project, and WRMK and MTW are co–principal investigators; they equally contributed to the overall protocol design. WRMK wrote the complete first draft of the paper with the assistance of MTW, SJS, and CBP. All the authors approved the final version of the manuscript.

Conflicts of Interest
None declared.
Multimedia Appendix 1
Recruitment flowchart leading to the sample of 203 participants.
[DOCX File, 22 KB-Multimedia Appendix 1]

Multimedia Appendix 2
Questionnaires administered across data collection visits.
[DOCX File, 16 KB-Multimedia Appendix 2]

Multimedia Appendix 3
Magnetic resonance imaging acquisition and preprocessing methods.
[DOCX File, 14 KB-Multimedia Appendix 3]

Multimedia Appendix 4
Descriptive statistics for income-to-needs ratios for the total sample and stratified by recruitment cell.
[DOCX File, 14 KB-Multimedia Appendix 4]

Multimedia Appendix 5
Descriptive statistics for bank savings for the total sample and stratified by recruitment cell.
[DOCX File, 14 KB-Multimedia Appendix 5]

Multimedia Appendix 6
Peer-review report from the National Institute of Child Health and Human Development - Psychosocial Development, Risk and Prevention (PDRP) Study Section - Risk, Prevention and Health Behavior Integrated Review Group - Center for Scientific Review (National Institutes of Health, USA).
[PDF File (Adobe PDF File), 154 KB-Multimedia Appendix 6]

References
1. Evans GW. The environment of childhood poverty. Am Psychol 2004;59(2):77-92. [doi: 10.1037/0003-066X.59.2.77] [Medline: 14992634]
2. Hackman DA, Farah MJ, Meaney MJ. Socioeconomic status and the brain: mechanistic insights from human and animal research. Nat Rev Neurosci 2010 Sep;11(9):651-659 [FREE Full text] [doi: 10.1038/nrn2897] [Medline: 20725096]
3. Noble KG, Houston SM, Brito NH, Bartsch H, Kan E, Kuperman JM, et al. Family income, parental education and brain structure in children and adolescents. Nat Neurosci 2015 May;18(5):773-778 [FREE Full text] [doi: 10.1038/nn.3983] [Medline: 25821911]
4. McLoyd VC. Children in poverty: development, public policy and practice. In: Damon W, Sigel IE, Renninger KA, editors. Handbook of Child Psychology: Child Psychology in Practice. 5th ed. Hoboken, NJ, USA: John Wiley & Sons; 1998:135-208.
5. Burchinal M, McCartney K, Steinberg L, Crosnoe R, Friedman SL, McLoyd V, NICHD Early Child Care Research Network. Examining the Black-White achievement gap among low-income children using the NICHD study of early child care and youth development. Child Dev 2011;82(5):1404-1420. [doi: 10.1111/j.1467-8624.2011.01620.x] [Medline: 21790543]
6. Duncan GI, Magnuson K, Kalil A, Ziol-Guest K. The importance of early childhood poverty. Soc Indic Res 2012;108(1):87-98. [doi: 10.1007/s11205-011-9867-9]
7. Noble KG, McCandliss BD, Farah MJ. Socioeconomic gradients predict individual differences in neurocognitive abilities. Dev Sci 2007 Jul;10(4):464-480. [doi: 10.1111/j.1467-7687.2007.00600.x] [Medline: 17552936]
8. Diamond A. Executive functions. Annu Rev Psychol 2013;64:135-168 [FREE Full text] [doi: 10.1146/annurev-psych-113011-143750] [Medline: 23020641]
9. Hanson JL, Chandra A, Wolfe BL, Pollak SD. Association between income and the hippocampus. PLoS One 2011 May 04;6(5):e18712 [FREE Full text] [doi: 10.1371/journal.pone.0018712] [Medline: 21573231]
10. Lipina SJ, Posner MI. The impact of poverty on the development of brain networks. Front Hum Neurosci 2012 Aug 17;6:238 [FREE Full text] [doi: 10.3389/fnhum.2012.00238] [Medline: 22912613]
11. Luby J, Belden A, Botteron K, Marrus N, Harms MP, Babb C, et al. The effects of poverty on childhood brain development: the mediating effect of caregiving and stressful life events. JAMA Pediatr 2013 Dec;167(12):1135-1142 [FREE Full text] [doi: 10.1001/jamapediatrics.2013.3139] [Medline: 24165922]
12. Noble KG, Houston SM, Kan E, Sowell ER. Neural correlates of socioeconomic status in the developing human brain. Dev Sci 2012 Jul;15(4):516-527 [FREE Full text] [doi: 10.1111/j.1467-7687.2012.01147.x] [Medline: 22709401]
13. Hair NL, Hanson JL, Wolfe BL, Pollak SD. Association of child poverty, brain development, and academic achievement. JAMA Pediatr 2015 Sep;169(9):822-829 [FREE Full text] [doi: 10.1001/jamapediatrics.2015.1475] [Medline: 26192216]

14. Camerota M, Willoughby MT. Applying interdisciplinary frameworks to study prenatal influences on child development. Child Dev Perspect 2021 Mar;15(1):24-30. [doi: 10.1111/cdep.12395]

15. Blair C. Developmental science and executive function. Curr Dir Psychol Sci 2016 Feb 01;25(1):3-7 [FREE Full text] [doi: 10.1177/0963721415622634] [Medline: 26985139]

16. Andersen SL, Carlé A, Karmisholt J, Pedersen IB, Andersen S. Mechanisms in endocrinology: neurodevelopmental disorders in children born to mothers with thyroid dysfunction: evidence of fetal programming? Eur J Endocrinol 2017 Jul;177(1):R27-R36. [doi: 10.1530/EJE-16-0947] [Medline: 28377377]

17. Girchenko P, Lahit-Pulkkinen M, Heikonen K, Reynolds RM, Laivuori H, Lipsanen J, et al. Persistently high levels of maternal antenatal inflammation are associated with and mediate the effect of prenatal environmental adversities on neurodevelopmental delay in the offspring. Biol Psychiatry 2020 May 15;87(10):898-907. [doi: 10.1016/j.biopsych.2019.12.004] [Medline: 31987493]

18. Sanchez CE, Barry C, Sabhlok A, Russell K, Kollins SH, et al. Maternal pre-pregnancy obesity and child neurodevelopmental outcomes: a meta-analysis. Obes Rev 2018 Apr;19(4):464-484 [FREE Full text] [doi: 10.1111/obr.12643] [Medline: 29164765]

19. Bale TL, Baram TZ, Brown AS, Goldstein JM, Insel TR, McCarthy MM, et al. Early life programming and neurodevelopmental disorders. Biol Psychiatry 2010 Aug 15;68(4):314-319 [FREE Full text] [doi: 10.1016/j.biopsych.2010.05.028] [Medline: 20674602]

20. Tau GZ, Peterson BS. Normal development of brain circuits. Neuropsychopharmacology 2010 Jan;35(1):147-168 [FREE Full text] [doi: 10.1038/npp.2009.115] [Medline: 19794405]

21. Buss C, Davis EP, Muftuler LT, Head K, Sandman CA. High pregnancy anxiety during mid-gestation is associated with decreased gray matter density in 6-9-year-old children. Psychoneuroendocrinology 2010 Jan;35(1):141-153 [FREE Full text] [doi: 10.1016/j.psyneuen.2009.07.010] [Medline: 19674845]

22. Brooks-Gunn J. The effects of poverty on children. Future Child 1997;7(2):55-71. [Medline: 9299837]

23. Holochwost SJ, Volpe VV, Gueron-Sela N, Propper CB, Mills-Koonce WR. Sociodemographic risk, parenting, and inhibitory control in early childhood: the role of respiratory sinus arrhythmia. J Child Psychol Psychiatry 2018 Sep;59(9):973-981 [FREE Full text] [doi: 10.1111/jcpp.12889] [Medline: 29532459]

24. Mills-Koonce WR, Willoughby MT, Garrett-Peters P, Wagner N, Vernon-Feagans L, Family Life Project Key Investigators. The interplay among socioeconomic status, household chaos, and parenting in the prediction of child conduct problems and callous-unemotional behaviors. Dev Psychopathol 2016 Aug;28(3):757-771 [FREE Full text] [doi: 10.1017/S0954579416000298] [Medline: 27427804]

25. Yu X, Quante M, Rueschman M, Ash T, Kaplan ER, Guo N, et al. Sleep 2021 Mar 12;44(3):zsaa193 [FREE Full text] [Medline: 33057653]

26. Golinkoff RM, Hoff E, Rowe ML, Tamis-LeMonda CS, Hirsh-Pasek K. Language matters: denying the existence of the 30-million-word-gap has serious consequences. Child Dev 2019 May;90(3):985-992. [doi: 10.1111/cdev.13128] [Medline: 30102419]

27. Sperry DE, Sperry LL, Miller PJ. Reexamining the verbal environments of children from different socioeconomic backgrounds. Child Dev 2019 Jul(94(4)):1303-1318. [doi: 10.1111/cdev.13072] [Medline: 29707767]

28. Bernier A, Carlson SM, Matte-Gagné C. Social factors in the development of early executive functioning: a closer look at the caregiving environment. Dev Sci 2012 Jan;15(1):12-24. [doi: 10.1111/j.1467-7687.2011.01093.x] [Medline: 22251288]

29. Blair C, Raver CC, Berry DJ, Family Life Project Investigators. Two approaches to estimating the effect of parenting on the development of executive function in early childhood. Dev Psychol 2014 Feb;50(2):554-565 [FREE Full text] [doi: 10.1037/a0033647] [Medline: 23834294]

30. Towe-Goodman NR, Willoughby M, Blair C, Gustafsson HC, Mills-Koonce WR, Cox MJ, Family Life Project Key Investigators. Fathers’ sensitive parenting and the development of early executive functioning. J Fam Psychol 2014 Dec;28(6):867-876 [FREE Full text] [doi: 10.1037/a0038128] [Medline: 25347539]

31. Sadeh A, Gruber R, Raviv A. Sleep, neurobehavioral functioning, and behavior problems in school-age children. Child Dev 2002;73(2):405-417. [doi: 10.1111/1467-8624.00414] [Medline: 11949899]

32. Bernier A, Carlson SM, Bordeleau S, Carrier J. Relations between physiological and cognitive regulatory systems: infant sleep regulation and subsequent executive functioning. Child Dev 2010;81(6):1739-1752. [doi: 10.1111/j.1467-8624.2010.01507.x] [Medline: 21077861]

33. Cajochen C, Knoblauch V, Kräuchi K, Renz C, Wirz-Justice A. Dynamics of frontal EEG activity, sleepiness and body temperature under high and low sleep pressure. Neuroreport 2001 Jul 20;12(10):2277-2281. [doi: 10.1097/00001756-200107200-00046] [Medline: 11447349]

34. Horne JA. Human sleep, sleep loss and behaviour. Implications for the prefrontal cortex and psychiatric disorder. Br J Psychiatry 1993 Mar;162;413-419. [doi: 10.1192/bjp.162.3.413] [Medline: 8453439]

35. Hughes CH, Ensor RA. How do families help or hinder the emergence of early executive function? New Dir Child Adolesc Dev 2009;2009(123):35-50. [doi: 10.1002/cd.234] [Medline: 19306273]
62. So K, Buckley P, Adamson TM, Horne RS. Actigraphy correctly predicts sleep behavior in infants who are younger than six months, when compared with polysomnography. Pediatr Res 2005 Oct;58(4):761-765. [doi: 10.1203/PDR.0000180568.97221.56] [Medline: 16189206]

63. Hall WA, Liva S, Moynihan M, Saunders R. A comparison of actigraphy and sleep diaries for infants' sleep behavior. Front Psychiatry 2015 Feb 12;6:19 [FREE Full text] [doi: 10.3389/fpsyt.2015.00019] [Medline: 25729371]

64. Hart B, Risley TR. Meaningful Differences in the Everyday Experience of Younger Children. Baltimore, MD, USA: Paul H Brookes Publishing; 1995.

65. Gilmore JH, Kang C, Evans DD, Wolfe HM, Smith JK, Lieberman JA, et al. Prenatal and neonatal brain structure and white matter maturation in children at high risk for schizophrenia. Am J Psychiatry 2010 Sep;167(9):1083-1091 [FREE Full text] [doi: 10.1176/appi.ajp.2010.09101492] [Medline: 20516153]

66. Gilmore JH, Shi F, Woolson SL, Knickmeyer RC, Short SJ, Lin W, et al. Longitudinal measurement of cortical and subcortical gray matter from birth to 2 years. Cereb Cortex 2012 Nov;22(11):2478-2485 [FREE Full text] [doi: 10.1093/cercor/bhr327] [Medline: 22109543]

67. Gilmore JH, Zhai G, Wilber K, Smith JK, Lin W, Gerg G. 3 Tesla magnetic resonance imaging of the brain in newborns. Psychiatry Res 2004 Nov 15;132(1):81-85. [doi: 10.1016/j.psychnres.2004.04.004] [Medline: 15546705]

68. Short SJ, Eliason JT, Goldman BD, Connelly M, Gu H, Shorte S, et al. Associations between white matter microstructure and infants' working memory. Neuroimage 2013 Jan 01;64:156-166 [FREE Full text] [doi: 10.1016/j.neuroimage.2012.09.021] [Medline: 22989623]

69. DWI/DTI Quality Control Tool: DTIPrep. NeuroImage Tools & Resources Collaboratory. 2022. URL: https://www.nitrc.or

70. Joshi S, Davis B, Jomier M, Gerig G. Unbiased diffeomorphic atlas construction for computational anatomy. Neuroimage 2004;23 Suppl 1:S151-S160. [doi: 10.1016/j.neuroimage.2004.07.068] [Medline: 15501084]

71. Smith SM. Fast robust automated brain extraction. Hum Brain Mapp 2002 Nov;17(3):143-155 [FREE Full text] [doi: 10.1002/hbmm.10062] [Medline: 12391568]

72. Yap PT, Gilmore JH, Lin W, Shen D. PopTract: population-based tractography. IEEE Trans Med Imaging 2011 Oct;30(10):1829-1840 [FREE Full text] [doi: 10.1109/TMI.2011.2154385] [Medline: 21571607]

73. Greicius MD, Srivastava G, Reiss AL, Menon V. Default-mode network activity distinguishes Alzheimer's disease from healthy elderly: evidence from functional MRI. Proc Natl Acad Sci U S A 2004 Mar 30;101(13):4637-4642 [FREE Full text] [doi: 10.1073/pnas.0308627101] [Medline: 15070770]

74. Zhu H, Kong L, Liu J, Stynner M, Gerg G, Lin W, et al. FADTTS: functional analysis of diffusion tensor tract statistics. Neuroimage 2009 Mar;45(1 Suppl):S133-S142 [FREE Full text] [doi: 10.1016/j.neuroimage.2008.10.060] [Medline: 19053945]

75. Calhoun VD, Adali T, Pearlson GD, Pekar JJ. A method for making group inferences from functional MRI data using independent component analysis. Hum Brain Mapp 2002 Nov;14(3):140-151 [FREE Full text] [doi: 10.1002/hbmm.1048] [Medline: 11559559]

76. Greicius MD, Srivastava G, Reiss AL, Menon V. Default-mode network activity distinguishes Alzheimer's disease from healthy elderly: evidence from functional MRI. Proc Natl Acad Sci U S A 2004 Mar 30;101(13):4637-4642 [FREE Full text] [doi: 10.1073/pnas.0308627101] [Medline: 15070770]

77. Calhoun VD, Adali T, Pearlson GD, Pekar JJ. A method for making group inferences from functional MRI data using independent component analysis. Hum Brain Mapp 2002 Nov;14(3):140-151 [FREE Full text] [doi: 10.1002/hbmm.1048] [Medline: 11559559]

78. Fox MD, Zhang D, Snyder AZ, Raichle ME. The global signal and observed anticorrelated resting state brain networks. J Neurophysiol 2009 Jun;101(6):3270-3283 [FREE Full text] [doi: 10.1152/jn.90777.2008] [Medline: 19339462]

79. Murphy K, Birn RM, Handwerker DA, Jones TB, Bandettini PA. The impact of global signal regression on resting state correlations: are anti-correlated networks introduced? Neuroimage 2009 Feb 01;44(3):893-905 [FREE Full text] [doi: 10.1016/j.neuroimage.2008.09.036] [Medline: 18976716]

80. Willoughby MT, Blair CB. Longitudinal measurement of executive function in preschoolers. In: Griffin JA, McCadle P, Freund LS, editors. Executive Function in Preschool Age Children: Integrating Measurement, Neurodevelopment and Translational Research. Washington, DC, USA: American Psychological Association; 2016:91-113.

81. Willoughby MT, Blair CB, Kuhn LJ, Magnus BE. The benefits of adding a brief measure of simple reaction time to the assessment of executive function skills in early childhood. J Exp Child Psychol 2018 Jun;170:30-44. [doi: 10.1016/j.jecp.2018.01.003] [Medline: 29407186]

82. Willoughby MT, Blair CB, Wirth RJ, Greenberg M. The measurement of executive function at age 5: psychometric properties and relationship to academic achievement. Psychol Assess 2012 Mar;24(1):226-239 [FREE Full text] [doi: 10.1037/a0025361] [Medline: 21966934]
85. Willoughby MT, Blair CB, Wirth RJ, Greenberg M. The measurement of executive function at age 3 years: psychometric properties and criterion validity of a new battery of tasks. Psychol Assess 2010 Jun;22(2):306-317. [doi: 10.1037/a0018708] [Medline: 20528058]
86. Smith-Donald R, Raver CC, Hayes T, Richardson B. Preliminary construct and concurrent validity of the preschool self-regulation assessment (PSRA) for field-based research. Early Child Res Q 2007;22(2):173-187. [doi: 10.1016/j.ecresq.2007.01.002]
87. Enders CK. Applied Missing Data Analysis. New York, NY, USA: The Guilford Press; 2010.
88. Schafer JL, Graham JW. Missing data: our view of the state of the art. Psychol Methods 2002 Jun;7(2):147-177. [Medline: 12090408]
89. Mackinnon DP, Lockwood CM, Williams J. Confidence limits for the indirect effect: distribution of the product and resampling methods. Multivariate Behav Res 2004 Jan 01;39(1):99 [FREE Full text] [doi: 10.1207/s15327906mbr3901_4] [Medline: 20157642]
90. Williams J, Mackinnon DP. Resampling and distribution of the product methods for testing indirect effects in complex models. Struct Equ Modeling 2008;15(1):23-51 [FREE Full text] [doi: 10.1080/10705510701758166] [Medline: 20179778]
91. Report on the economic well-being of U.S. households in 2018 - May 2019. U.S. Board of Governors of the Federal Reserve System. 2013. URL: https://www.federalreserve.gov/publications/2019-economic-well-being-of-us-households-in-2018-dealing-with-unexpected-expenses.htm [accessed 2022-05-09]

Abbreviations
- **BEE**: Brain and Early Experience
- **CV**: coefficient of variation
- **DTI**: diffusion tensor imaging
- **EF**: executive function
- **fMRI**: functional magnetic resonance imaging
- **ITN**: income-to-needs
- **LENA**: Language Environment Analysis
- **rs**: resting state
- **SES**: socioeconomic status
- **UNC**: University of North Carolina

©William Roger Mills-Koonce, Michael T Willoughby, Sarah J Short, Cathi B Propper. Originally published in JMIR Research Protocols (https://www.researchprotocols.org), 29.06.2022. This is an open-access article distributed under the terms of the Creative Commons Attribution License (https://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work, first published in JMIR Research Protocols, is properly cited. The complete bibliographic information, a link to the original publication on https://www.researchprotocols.org, as well as this copyright and license information must be included.