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Lindsey S. Rudov April 18, 2012

Prenatal Exposure to Maternal Distress and Child Behavior During the Preschool Years

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Abstract

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This study investigated the relationship between prenatal maternal distress and preschool children's internalizing and externalizing problems. This study also evaluated potential mediating (stress reactivity) and moderating (postnatal stress exposure) factors related to this process, thereby adding to the literature a clearer picture of the risk pathway associated with prenatal maternal distress exposure. During a lab visit, 74 preschool aged children were exposed to two consecutive stressor tasks, and baseline and post-stressor cortisol levels were analyzed in order to investigate HPA axis dysregulation. Child behavior was measured using a standardized assessment and was completed by both the mother and an alternate caregiver in order to reduce the impact of maternal bias on the results. Results revealed that prenatal maternal distress exposure was associated with HPA axis dysregulation in this sample and that child cortisol reactivity mediated the relationship between prenatal maternal distress and externalizing problems in childhood. Specifically, cortisol reactivity assumed a key role in the development of aggressive behavior in childhood; however, it did not mediate the relationship between prenatal maternal distress and child attention problems. Moreover, results suggested a moderating effect of current maternal stress on the relationship between child cortisol reactivity and behavior problems. Results indicated that greater levels of cortisol reactivity are more likely to be associated with higher levels of behavioral problems in children if the child is exposed to recent stressful life events. An interpretation of these findings and implications for clinical practice are addressed.

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Children of mothers who suffer from mental illness are at risk for psychopathology due to both genetic factors and postnatal environmental factors (such as increased exposure to stress). Importantly, maternal disorders that occur during the prenatal stage of development may also have additional harmful environmental effects on the offspring. Previous research has demonstrated an association between maternal prenatal distress and cognitive and behavioral problems in children (Brand & Brennan, 2009). What is less well understood are the risk pathways that link prenatal distress to negative child outcomes, and the mediators and moderators that influence these risk pathways. The present study is designed to examine the association between prenatal maternal distress and child psychopathology, and specifically to test the role of cortisol reactivity as a mediator and postnatal stress exposure as a moderator in this process.

Theoretical Foundation

The importance of the fetal environment for future development has been described by the theory of *fetal programming*, originally proposed by Dr. David Barker in 1989 (The Barker Hypothesis). Barker suggested that the environment in which the fetus develops is the environment that the fetus anticipates that it will be required to adapt to, after birth and throughout development. Problems arise when there is a mismatch between the prenatal and postnatal environments in which an individual develops because the individual is equipped to adapt most optimally in a context similar to his or her fetal environment. The majority of research that aims to examine the Fetal Programming Theory has been conducted in order to assess physical health outcomes associated with the prenatal environment. This is because Barker's initial work determined that the prenatal environment had a strong impact on the offspring's potential development of chronic disease later in life. Due to consistent findings that

link prenatal conditions to physiological outcomes (Ellison, 2010), researchers have recently become interested in the application of the fetal programming theory to psychological and psychiatric outcomes as well (Coe & Lubach, 2008).

Initial research investigating the effects of the prenatal environment on psychological and psychiatric outcomes noted a relationship between factors reflective of maternal food intake (e.g. pregnancy during a famine or low birth weight) and a higher likelihood of offspring mood disorders or psychotic disorders (Ellison, 2010). More recently, the field has begun to investigate the impact of a more elusive determinant of the prenatal environment: maternal stress. Maternal experience of stress during pregnancy has the potential to influence a multitude of the mother's physiological systems that have a direct impact on the fetal environment. For instance, physiological activity such as placental functioning, nutrient transfer, and endocrine activity may all have a substantial influence on the development of brain functioning, including neuromotor maturation, emotion reactivity, and the activity of the hypothalamic-pituitary-adrenal (HPA) Axis (Coe & Lubach, 2008). Work that initially focused on maternal stress exposure has since been broadened to encompass other factors of maternal distress such as depression and anxiety. A recent review concluded that these aspects of maternal distress (stress, depression, and anxiety) are highly correlated with one another during pregnancy and appear to have similar effects on child behavioral outcomes (Brand & Brennan, 2009). The current study, therefore, will utilize a combined measure of maternal distress, consisting of maternal anxiety, depression, and perceived distress during pregnancy as it relates to child behavior outcomes in the preschool period.

Prenatal Maternal Distress Exposure and Child Behavioral Outcomes

Negative child outcomes associated with maternal prenatal distress range from temperament problems in infancy, such as greater irritability (Monk, 2001), to behavioral

problems in childhood such as deficiencies in motor activity and learning (DiPietro, 2004). Although it is also possible to study associations between prenatal maternal distress and developmental outcomes in adolescence and adulthood, it is most optimal to study associations in infancy and childhood due to the likelihood of fewer confounds (such as puberty, peer influences, etc.) during these early years.

When studying the effects of the prenatal environment on child development, an initial area of interest is neurobehavioral outcomes. By examining the neurobehavioral development of a child exposed to prenatal maternal distress, researchers have the ability to gain an understanding about how fetal environmental factors affect brain development during gestation. For instance, in a 2003 study conducted by Coe and colleagues, it was found that prenatal stress exposure negatively affected volume of the hippocampus and the number of glucocorticoid receptors in the hippocampus of the offspring (as cited in Glover, O'Connor, & O'Donnell, 2010). Thus, exposure to prenatal maternal distress can have profound effects on the development of the fetal brain, and as discussed later in this thesis, such insults can lead to psychopathology in the child.

A review of the past twenty years of prospective studies examining the effects of prenatal exposure to maternal distress and associated neurobehavioral development of the fetus concludes that prenatal maternal distress is related to regulation problems at emotional, behavioral, and cognitive levels (Van den Bergh, Mulder, Mennes, & Glover, 2005). In this review, the authors note that all six of the prospective studies that have investigated preschool children have found that prenatal exposure to maternal distress is linked to behavior problems during these years. In particular, problems of poor attention and hyperactivity are reoccurring behavioral problems that children exposed to prenatal maternal distress have displayed (Van den Bergh et al., 2005).

Huizink, Buitelaar, and Mulder (2004) conducted a meta-analysis of the behavioral consequences of offspring exposed to prenatal stress for rodents and non-human primates. The impaired behavior of rodents that may be analogous to humans includes: “less activity”, “reduced propensity for social interaction”, and “more emotionality in novel situations” (Huizink et al., 2004). In reference to non-human primates, research demonstrates that the offspring may develop behavioral impairments in responses to novelty and social interaction such as “fewer social interactions and withdrawal from social interactions” and “more disturbance behavior” (Huizink et al., 2004). From this meta-analysis, however, it is evident that there is no consensus on the definition and quantitative standard of “prenatal stress”. Furthermore, due to different methodologies used by different investigators, the data are not entirely conclusive (Huizink et al., 2004). Although animal studies are a useful pre-requisite to studies involving humans – and are easier for researchers to conduct – the prenatal stress that these organisms are exposed to differs greatly from the maternal prenatal distress a human fetus may be exposed to, and animal studies must only be used as a guide in the formation of future studies with humans.

There is a relative lack of human research investigating the relationship, however, between prenatal exposure to maternal distress and the behavior of the child past the infancy stage. As described above, only six prospective studies have been conducted to date (Van den Bergh et al., 2005), and it is necessary to explore this association further due to the paucity of research. Moreover, the majority of the research connecting prenatal exposure to maternal distress and behavior during the preschool period has relied solely on measures completed by the mother, which have the high potential of being confounded or biased (Field, 2002). The current study will use multiple measures of maternal distress, including self-reports, interviewer reports, and clinical judgments, to attempt to minimize the impact of bias in maternal reports on outcomes.

Another unanswered question in this area of research is whether maternal prenatal distress is relevant to the prediction of internalizing problems, externalizing problems, or both. For instance, longitudinal data regarding the development of monkeys demonstrates that those exposed to prenatal stress demonstrated "delayed motor development and reduced attention in infancy" followed by negative behaviors and "dysfunctional social behavior with peers" that lasted through adolescence (Schneider & Moore, 2002, as cited in DiPietro, 2004). Moreover, other research concludes that children born to depressed mothers show "dysregulated arousal and attention" (Field, 2002), and there is evidence demonstrating an association between maternal depression and externalizing problems, such as behavioral disruption and attention problems (Goodman, 2007). It is necessary that the future direction of this research examine what types of behaviors can result from prenatal maternal distress in order to better identify the risk pathway of psychopathological development for this high-risk group.

Two other issues of central importance to this thesis are the mediating and moderating factors that might impact the risk pathway from maternal prenatal distress to child psychopathology. Specifically, we are interested in the potentially mediating effects of the HPA axis and the moderating effects of postnatal exposure to stress. More information concerning mediators will help in guiding theory development, whereas more information concerning moderator effects might be useful in planning targeted intervention or prevention programs for individuals at particularly high risk for psychopathology.

The HPA Axis as a Mediator of the Risk Pathway

The examination of mediators allows researchers to assess whether certain mechanisms are partly responsible for the association between the predictor and outcome variables in question. The present study will explore the potential mediating role of the HPA axis in the association between prenatal maternal distress and child psychopathology.

The HPA axis is a physiologically important system that involves both neural and endocrine subsystems, and plays a primary role in both mental and physical responses in the context of stress. Because the three different elements of the HPA axis (the hypothalamus, the pituitary gland, and the adrenal glands) begin to develop during gestation, the mother's state of being during pregnancy can be highly influential to their development. For example, pregnant mothers themselves with mood disorders have a dysregulated HPA axis, and therefore provide a non-optimal fetal environment for their offspring due to the negative influence of maternal stress hormones on the placenta and the developing fetus (Huizink et al., 2004). Seckl and Meaney (2004) have studied how fetal programming can occur in response to the exposure to glucocorticioids and ascertain that when a fetus is exposed to high levels of maternal stress hormones, this negatively impacts the development of the brain (Seckl & Meany, 2004).

In *Prenatal Stress and the Programming of the HPA Axis* (2010), Glover and colleagues discuss the array of research suggesting that prenatal stress exposure changes the functionality of the offspring's HPA axis. An ample amount of animal research conducted with a range of species demonstrates that when exposed to a "great intensity" of prenatal stress, the HPA axis of the offspring will develop in a sub-optimal manner (Glover et al., 2010). It should be noted, however, that the forms of prenatal stress that animals have been exposed to are delivered experimentally, and therefore these conclusions are not applicable to human fetuses who are exposed to prenatal maternal distress in a non-controlled fashion.

Although fewer in quantity, human studies show evidence for a relationship between HPA axis functionality and prenatal exposure to maternal distress as well (Glover et al., 2010). In particular, it is proposed that a fetus exposed to prenatal distress is simultaneously suffering from an overexposure to maternal glucocorticoids, and that this exposure programs the development of the child's HPA axis. Moreover, the insults caused by this overexposure have the

potential to impact the child's supply and regulation of his or her glucocorticoids (Seckl & Meaney, 2004), which results in dysregulated activity of the HPA axis and further risk for child psychopathology.

When an individual attempts to cope with stress, his or her HPA axis releases a series of regulating hormones that the body uses to return itself to homeostasis in response to the stressor. Researchers are able to measure these stress hormones and draw conclusions about the activity and regulatory capabilities of one's HPA axis. Of great popularity in the literature has been the measurement of the stress hormone cortisol, for it is easily obtainable via both plasma and salivary samples. Numerous studies have concluded that children exposed to maternal prenatal distress evidence abnormal outputs of cortisol in response to stress, elucidating the negative effect that prenatal distress will have on the child's HPA axis (Brand & Brennan, 2009; Gutteling, de Weerth, & Buitelaar, 2005; Huizink et al., 2004). Moreover, research demonstrates that higher levels of corticotropin (ACTH), which is the hormone that triggers the production of cortisol, are present in children prenatally exposed to maternal stress (Huizink et al., 2004; Glover et al., 2010). The majority of the research regarding stress hormones in high-risk populations, however, has investigated the hormonal outputs of infants or adults, and research measuring cortisol in high-risk children during the preschool years is lacking. Thus, the current study adds to the literature by analyzing HPA axis activity of preschoolers exposed to prenatal maternal distress.

Although existing research demonstrates a significant difference between cortisol outputs of those children prenatally exposed to maternal distress compared to other children, there is no general consensus in the literature regarding whether lower or higher basal cortisol levels as compared to the normal population are most detrimental (Sondeijker, Ferdinand, Oldehinkel, Timeier, Ormel, & Verhulst, 2008). The discrepancy regarding results of basal cortisol levels

(Sondeijker et al., 2008) has led researchers to view *cortisol reactivity* as the more optimal measurement of the HPA axis. By quantifying how an individual's level of cortisol changes in response to a stressor, Schlotz and Phillips (2009) claim that this reactivity can represent "the extent to which a person is likely to respond to a stressful event". Because of this research, the current study will operationalize HPA axis activity as one's cortisol *reactivity*, rather than one's basal or diurnal levels of cortisol.

The precursors leading to HPA axis dysregulation (e.g. prenatal maternal stress) are of great interest to researchers in part because of the associated mental health problems this HPA axis dysregulation can lead to, such as delinquency, substance abuse (Sondeijker et al., 2008), and other forms of psychopathology. In fact, ample research demonstrates an association between dyregulation of the HPA axis (as measured by cortisol) and behavioral problems in childhood, such as an increase in anxiety-related behaviors and problems of attention, disruption, and aggression (Goodman, 2007; Glover et al., 2010). In summary, current research shows that children who evidence a dysregulated HPA axis are more susceptible to the development of externalizing behaviors in childhood. Because prenatal distress has also been linked to disruptions in HPA axis functioning, this study will examine HPA axis dysregulation, and specifically cortisol reactivity, as a potential mediator between maternal prenatal distress and preschool age child behavior problems.

The Moderating Effect of Postnatal Stress Exposure

Although a substantial amount of research demonstrates that maternal prenatal distress may be associated with the development of child psychopathology, the offspring's *postnatal* environment must be considered in order to fully understand the developmental trajectory this child experiences. Children who are already at high-risk for the development of psychopathology (such as those in the current sample) may be at an even greater risk if exposed to high levels of

stressors in their postnatal environment. As described previously, the HPA axis is an important mechanism that is responsible for the individual's response to and control of stress. Therefore, the current study also predicts that the child's postnatal environment – as created by his or her mother – will have a moderating effect on the relationship between the child's HPA axis dysregularity and his or her behavioral problems. In other words, it is hypothesized that children with HPA axis dysregularity who are *also* exposed to higher levels of postnatal stress are the children who are at most risk for the development of behavioral problems in childhood.

Because most of the toddler and pre-schooler's post-natal environment is constructed by the mother's daily activities and moods, it is relevant to study the mother's daily life for a general idea of the postnatal stress that her pre-school child is exposed to. As proposed by Hammen (2002), mothers who are depressed generate their own life stress as a result of their mental state (as cited in Goodman, 2007). Because mothers with past or current depression (which is a characteristic of our sample) may create exceptionally stressful environments for their offspring, it is predicted that these children are placed at an even greater risk of psychopathology due to their stressful postnatal environment.

Compelling research completed by Costello and colleagues (2007) suggests that postnatal stress exposure may serve as a moderator between prenatal distress and psychopathology. These researchers found that out of a sample of 1400 boys and girls, the girls who were vulnerable to an abnormal development of the HPA axis (due to a low birth weight) evidenced higher rates of depression than those who were born with a normal birth weight. What was groundbreaking about this research, however, was that with each additional postnatal stressor experienced (in the perinatal through adolescent periods), the low birth weight child's likelihood of displaying symptoms of psychopathology (namely depression) increased substantially (Ellison, 2010). This research suggests, therefore, that postnatal stress exposure may be of particularly important

influence for children who are born with an already existent vulnerability (such as exposure to prenatal maternal distress).

As Costello et al.'s study demonstrates, an individual who is inherently at risk for psychopathology may be at an even *greater* risk if he or she is exposed to additional life stress. This concept is referred to in the psychology literature as the Diathesis-Stress Model (Mash & Wolfe, 2010). In the current study, it is hypothesized that a dysregulated HPA axis will provide a *diathesis*, or internal susceptibility, toward the development of child psychopathology, and that when coupled with *stress* in the postnatal environment, the child will be at an even greater risk of psychopathology.

The Current Study

The current study will add to the literature by analyzing the moderating effect of maternal life stress on the relationship between a child's dysregulated HPA axis and behavioral problems during the preschool years. By investigating whether postnatal stress moderates the relationship between an already dysfunctional HPA axis and the development of certain behaviors, researchers will be better able to target factors for prevention in high-risk children. The current study also examines cortisol reactivity as a mediator between a child's exposure to prenatal maternal distress and his or her later behavior problems, and therefore adds to the literature by exploring a potential mechanism behind the effect. Increased understanding of mechanisms can aid in both theory development and intervention planning.

The current study also adds to the literature by utilizing a comprehensive and prospective variable of maternal distress during pregnancy. By coupling several different prenatal measures collected from different sources, the current study will more validly and reliably measure the mother's distress during pregnancy. Moreover, the current study investigates both internalizing and externalizing behaviors. By differentiating between these two behavior categories, we will

be better able to assess specific behavioral tendencies that are associated with prenatal maternal distress.

Clarification of Central Aims

This thesis includes two central aims: (1) to explore the risk pathway of children at high-risk for the development of psychopathology due to exposure to prenatal stress, and (2) to investigate how a stressful postnatal environment may moderate the relationship between HPA axis vulnerability and psychopathology. In order to evaluate these two central aims, the current study hypothesizes that children who are exposed to prenatal maternal stress will show greater behavioral problems in childhood, and that this relationship will be mediated by dysregulation of the child's HPA axis and moderated by exposure to postnatal stress (see Figure 1).

Method

Participants

The participants for this study were 74 children (n=74) and their mothers who took part in an NIMH funded study of outcomes associated with prenatal medication use (Smith and Brennan, PIs). Participants were included in the current study if: (a) prospective measures of maternal distress were collected during pregnancy and (b) both pre- and post-stressor salivary cortisol samples had been collected and assayed. In this group were 42 females and 32 males, ranging from the ages of 29.6 months (approximately 2 and a half years) to 67.6 months (approximately 5 and a half years). The mean age of the group was 47.5 months (approximately 4 years) (SD = 9.7 months).

Regarding the mothers of the child participants, 56 of the mothers were currently in psychiatric treatment during the study, 17 of the mothers were not currently in treatment during the study, and 1 mother's treatment status was unknown. Of these mothers, 30 completed graduate/professional school, 6 completed part of graduate/professional school, 22 graduated

from a 4-year college, 6 graduated from a 2-year college, 9 graduated part of college, and 1 was unknown. The majority of the sample was Caucasian (59), and 7 mothers were African Americans, 3 were Asian, 4 were Hispanic, and the ethnicity of 1 mother was unknown.

Procedure

Prenatal Maternal Distress Data Collection. Mothers included in the current study completed measures of distress during each trimester of pregnancy at the Women's Mental Health Program (WMHP) of Emory University. Because these distress measures were completed prospectively and at several points throughout the mother's pregnancy, the latent variable of prenatal maternal distress provided a more comprehensive picture of prenatal distress exposure than methodologies used by other studies.

Salivary Cortisol Collection. In order to obtain a measurement of stress reactivity, researchers collected a pre- and a post-stressor test salivary cortisol sample from the child. The first salivary cortisol sample was collected immediately upon entering the lab between the approximate hours of 9:30 am and 10:30 am. The first collection was collected immediately in order to retrieve the most valid baseline measurement of cortisol possible and to prevent any situational fear or anxiety that the child might have experienced in the lab environment from biasing the "baseline" measurement. It must be kept in mind, however, that the "baseline" measurement may be potentially confounded by the anticipation that the child experiences after entering a novel environment such as the laboratory.

To obtain the saliva sample, the child chewed on a 2-inch piece of cotton dental roll for approximately 1 minute in order to moisten the roll with saliva. The roll was dipped in 1/64th teaspoon of cherry Kool-Aid to improve the experience for the child. After the roll absorbed enough saliva, the researcher placed the saliva-coated roll into a plastic syringe. All researchers wore rubber gloves in order to prevent contamination. After placed properly in the syringe, the

researcher drained the saliva from the dental roll into a plastic vial. The vial was then properly labeled with participant ID, date, and time of collection. The vial was then placed in a -20 degree Celsius freezer. The identical protocol was executed for retrieval of the post-stressor saliva sample.

Stressor Tasks. Two consecutive stressor tasks were completed. First, the child completed the Clear Box Task, which lasted for two minutes. Before the timer started, the child was asked by the researcher to pick one of two desired toys: either a slinky or a bouncy ball. After selecting the desired toy, the researcher placed the toy into a 7.5" x 7.5" transparent plastic box with a hinged lid and clasp. All of these actions took place in front of the child in order to entice him/her, and the child was not allowed to touch the desired toy. The researcher then placed a padlock onto the clear box hinge and handed the child a small ring of keys. Although none of the keys opened the padlock, the child was instructed to open the padlock with one of the keys in order to retrieve the desired toy, and was also told that "most kids do this pretty fast". Upon the researcher's exit of the room, the video recording started, marking the beginning of the two minutes of the task. The researcher then returned to the video recording room in order to watch the child's performance during the task and remain alert to any problems.

While in the video recording room, the researcher watched the child's behavior in response to the stressor task. The researcher rated the child's level of frustration during the stressor task on a likert scale of 1 to 5 (1 being the least frustrated and 5 being the most frustrated). A Frustration Rating of 1 was given if the child was not at all frustrated (e.g. shows little affect, completes task with ease, etc.). A Frustration Rating of 5 was given if the child was highly frustrated/distressed (e.g. showed multiple signs of negative affect, refused to continue, became visibly upset, etc.)

After two minutes, the researcher returned to the testing room and apologized for not giving the child the correct key. The child was then given the desired toy to play with while the researcher placed the materials of the Clear Box task above the cabinet in the room and retrieved materials for the following stressor task, which would last 3 minutes.

For the second stressor task, Green Circles, the researcher sat herself next to the child at the table and placed one sheet of 11x17" white paper and a Ziploc bag of broken crayons in front of the child. The child was told to "draw a perfect green circle" and the researcher then began a stopwatch out of the child's view. After the child drew his/her first green circle, the researcher critiqued it by saying "That circle is too small/skinny/big/pointy, etc. Draw me another one". After the second green circle was drawn and critiqued, the researcher stood up and taped two examples of "perfect" green circles to the cabinet and said, "The last two kids that were here drew these circles and they are perfect. I need you to draw a perfect circle. The other kids thought this was easy". Regardless of the manner in which the child responded throughout the task, the researcher maintained a neutral expression and three different times throughout the 3-minute task reiterated that she needed a "perfect" green circle.

The procedure was stopped after three minutes and the researcher reassured the child after his/her last attempt by saying, "That circle looks really good! Circles are hard to draw. Let's go show your mom what a great job you did!" The researcher gave the child a frustration rating after the Green Circles task by using the same coding criteria as the Clear Box task. The researcher then waited 15-20 minutes (the amount of time it takes for cortisol to transport to saliva) and obtained the post-stressor saliva sample. The child engaged in free play during this time. As she did with the first cortisol sample, the researcher immediately labeled the second saliva sample and placed it in the freezer.

Child Behavior and Post-natal Environment Data Collection. While the child completed both stressor tasks and other tasks not included in the current study (e.g. cognitive tasks, executive functioning tasks, etc.), the mother was asked to complete several additional questionnaires. During this time, the mother completed assessments that were reflective of the child's behavior and the child's post-natal environment. These measures are discussed in detail below. In addition, an alternate caregiver (such as a grandmother, teacher, or father) completed the same behavioral questionnaire and this questionnaire was either mailed into the lab or brought to the lab by the mother during the scheduled lab visit.

Measures

Prenatal Maternal Distress. Prenatal maternal distress was measured by combining data from four separate prospective measures that were completed during pregnancy. Two of these measures were self-reports, whereas the other two were completed by a research assistant or clinician. All four measures were completed at several different times during pregnancy (once during each trimester), and the current study therefore accounts for maternal distress throughout all trimesters. Scores from all prenatal administrations of the measurements were combined into four independent cumulative distress ratings by calculating an "area under the curve" (AUC) score for each distress measure.

AUC measures of the four prenatal distress measures were highly correlated and therefore the construction of a composite prenatal maternal distress score during pregnancy was undertaken. Using Structural Equation Modeling (SEM), a latent variable was created that combined these four observed measures (see Figure 2). By creating a latent variable, this study reduced the probability of error that would result from using only one measure of maternal distress. Moreover, by calculating "area under the curve" for each measure, the current study was

able to account for cumulative distress throughout the entire pregnancy. The latent variable included the following measures:

1. Hamilton Rating Scale for Depression (HRSD) (Hamilton, 1960)
2. Clinical Global Index (CGI) (Guy, 1976)
3. Beck Depression Inventory (BDI) (Beck, Steer, & Brown, 1997)
4. Perceived Stress Scale (PSS) (Cohen, Kamarck, & Mermelstein, 1983)

The particular types of distress (e.g. stress vs. depression symptoms) that the questionnaire aims to measure and the structure of each questionnaire are described in Table 1.

HPA Axis Dysregulation. In order to operationalize the activity of the child's HPA axis in response to the stressor tasks, researchers collected two cortisol samples: (a) a baseline measurement collected after entering the lab and (b) a post-stress measurement obtained 15-20 minutes after the completion of laboratory stressor tasks.

After collection and proper storage, saliva samples were sent to Yerkes Primate Center in order for concentration levels of cortisol to be extracted from the saliva. This process was completed on December 12, 2011 by using a commercially prepared kit produced by Salimetrics (State College, PA). Sensitivity for salivary cortisol is less than 0.003 µg/dL (Salimetrics, 2012). Inter-assay coefficients of variation were 10.83% at 0.97 µg/dL and 5.84% at 0.099 µg/dL. Intra-assay coefficients of variation were 9.82% at 0.97 µg/dL and 11.6% at 0.099 µg/dL.

HPA axis dysregulation was defined by higher levels of stress reactivity, which was calculated as the change in the child's cortisol levels from before to after the stressor tasks. The child's change in cortisol reflected his or her physiological and hormonal HPA axis activity. Cortisol reactivity scores were then analyzed as a mediator according to the proposed model (shown in Figure 1).

Child Behavior Checklist (CBCL) (Achenbach, 1991). The CBCL assesses the level of behavioral problems in children ages 1.5-5 years in the past 2 months. The questionnaire includes 99 behavioral descriptors and the rater assigns a 0 (not true), 1 (sometimes true), or 2 (very true) according to how well the descriptor corresponds to the child's general level of behavioral functioning over the last 60 days. Higher scores indicate higher ratings of behavior problems. The questionnaire yields an overall Total Problem score, an overall Internalizing Problem score, and an overall Externalizing Problem score. Additional scores are given in regard to more distinct behaviors (e.g. attention problems, withdrawn behavior, etc.). In the current study, both mothers and alternative caregivers (e.g. father, teacher, grandmother) completed the CBCL. Raw scores were used for data analyses.

Life Experiences Survey (LES) (Sarason Johnson, & Siegel, 1978). The LES was used to quantify the number of life events that a mother encountered in the past six months. Mothers also rate each life event that they have encountered based on how much stress that event caused for them. Ratings range from -3 (extremely negative) to 3 (extremely positive). Examples of life events included in the LES are: marriage, change in religious activities, trouble with the in-laws, traffic violations, loss of a loved one, change in employment situation, etc. The total number of stressful life events that the mother has encountered is calculated, as is the sum of the stress ratings.

Results

Data Analysis

Structural Equation Modeling. The current study used structural equation modeling (SEM) as the main statistical approach for data analysis. This approach was optimal due to the desired construction of a latent variable of prenatal maternal distress. Kline (2011) clarifies that latent variables are useful in data analysis because they afford one unified variable based on a

hypothetical construct that is not directly observable or quantifiable, but rather reflective of a ”true” value of the construct. Before creating the latent variable of prenatal maternal distress, correlation statistics were obtained for the four independent AUC measures obtained from the mothers during pregnancy. These measures were highly correlated (see Table 2), and therefore construction of a latent variable was performed.

A software program entitled AMOS 17.0 (Arbuckle, 2008) was used to assess relationships between prenatal maternal distress and cortisol reactivity and behavior problems in children. AMOS is able to predict parameter estimates of the population by using a method of maximum likelihood (ML), which is a statistical principle that produces parameter estimates “that maximize the likelihood (the continuous generalization) that the data (the observed covariances) were drawn from this population” (Kline, 2011, p. 154).

It must be kept in mind that SEM does not indicate causation, but this statistical tool does have the ability to confirm the consistency or inconsistency of the sample data as a representation of the specified model (Kline, 2011). The given hypothesis is either accepted or rejected based on several fit statistics that AMOS calculates as a measure of the fit between the sample and the parameter statistics. The current study evaluated three different measurements of “good fit”: the chi-square index, the comparative-based fit index (CFI), and the root-mean-square error of approximation (RMSEA). These fit statistics are reported for each model that was tested.

A non-significant chi-square test statistic demonstrates that the two models are not significantly different from each other, and therefore indicate that the observed sample data fit the hypothesized model. The CFI tests the fit between the sample model and the hypothesized model on a scale from 0 to 1. Values of .90 or higher are desirable and reflect good fit (Hu & Bentler, 1999). The RMSEA statistic, on the other hand, tests the lack of fit between the sample model and population model covariation matrix and also produces values on a scale of 0 to 1.

Values less than .06 reflect a good fit between the models (Kline, 2011). The present study used SEM to test the main effect hypothesis that maternal distress exposure throughout pregnancy would be positively associated with child behavioral problems. The present study also used SEM to test the mediational role of HPA axis dysregulation. The moderating role of the post-natal environment was evaluated by using multiple regression analyses conducted in SPSS (discussed below).

Testing for Mediation. In order to assess HPA axis dysregulation as a mediator, the association between prenatal maternal distress and child behavioral problems was first examined. Next a model was tested that included cortisol reactivity as a mediator between prenatal distress and child behavior outcomes. The mediational effect of cortisol reactivity was examined by comparing the Beta (regression) weight of the direct path between prenatal distress and the behavioral outcome to the Beta weight of the same path after controlling for cortisol reactivity. If the Beta statistic was no longer significant after controlling for cortisol reactivity, then the variable qualified as a mediator of the association.

Testing for Moderation. Multiple regression analyses were used in order to test the moderating effect of post-natal stress on the relationship between cortisol reactivity and child behavioral problems. These analyses were conducted using version 19 of SPSS. All main effect variables were mean centered before analyses. Two moderator variables were assessed: (a) total number of life events that the mother has experienced in the past six months, and (b) total [positive or negative] rating that the mother assigned to the major life events of the past six months. Interaction terms were created by multiplying cortisol reactivity by each of the LES moderators. Covariates and main effects were entered into the first blocks of a hierarchical regression analysis. Interaction terms were then entered into the third block, and F change statistics were evaluated to determine whether moderator effects were significant.

Assessing Potential Confounds

Bivariate correlations were examined between potential confounds and the dependent variables of internalizing and externalizing behaviors (reported by both maternal and alternate caregiver on the CBCL). Sub-categories of externalizing behaviors such as aggression and attention were also analyzed as exploratory dependent variables in the current study. As shown in Table 3, potential confounds such as child sex, child age, self reported maternal depression (Beck Depression Inventory; BDI) on day of the visit, total duration of maternal psychiatric illness (in months and across all diagnoses), mother's age, and mother's education were all examined as potential confounds. As can be seen in Table 3, current maternal BDI was the only variable that correlated with dependent measures (i.e. mother reported external, internal, and aggressive behavior raw scores). This confound was therefore controlled in all subsequent analyses. In addition, baseline cortisol was controlled for due to the potentially confounding effect of baseline cortisol on cortisol reactivity levels. Moderator analyses were also undertaken with controls for child illness on the day of the visit and recent tooth loss because these variables are two additional potential confounds of cortisol reactivity. These variables were excluded from SEM models because they negatively impacted model fit statistics. Finally, maternal medication use during pregnancy was assessed as a potential confound, but it was not found to be related to any of the dependent measures in the study.

Structural Equation Modeling

The descriptive statistics (e.g. minimum, maximum, mean, and standard deviation) of all variables discussed in this section are presented in Table 4.

Externalizing and Internalizing Behavior Problems – Mother Rated. Structural equation modeling was used in order to evaluate the relationship between prenatal maternal distress and child behavioral problems as indicated by the mother's CBCL ratings. Due to results of the

preliminary analyses, maternal BDI was originally controlled for in the model. When controlling for BDI, however, the model did not achieve good fit. Because test statistics revealed that the BDI was not predictive of the outcome variables (both externalizing and internalizing child behavior problems in the SEM model), maternal BDI was removed. After the removal of maternal BDI, a good fit was achieved for structural models predicting both internalizing and externalizing behavior problems as reported by the CBCL (internalizing: χ^2 (df = 5) = 3.92, p = .56, CFI = 1.00, RMSEA = .00; externalizing: χ^2 (df = 5) = 2.36, p = .80, CFI = 1.00, RMSEA = .00). Results indicated, however, that prenatal maternal distress did not significantly predict mother reported internalizing or externalizing behavior problems in children (internalizing: β = .07, p = .59; externalizing: β = .15, p = .21).

Externalizing and Internalizing Behavior Problems – Alternate Caregiver Rated. Next, we examined the relationship between prenatal distress exposure and externalizing and internalizing problems as rated by an alternate caregiver. The structural models predicting both internalizing and externalizing problems as rated by an alternate caregiver fit well (internalizing: χ^2 (df = 5) = 3.89, p = .57, CFI = 1.00, RMSEA = .00; externalizing: χ^2 (df = 5) = 1.96, p = .86, CFI = 1.00, RMSEA = .00). Results indicated that prenatal maternal distress did not significantly predict internalizing behavior problems as rated by an alternate caregiver (β = .13, p = .35). Prenatal maternal distress did, however, significantly predict externalizing behavior problems of the child as rated by an alternate caregiver (β = .31, p = .02). This model is shown in Figure 2.

Impact of Child Stress Reactivity on Externalizing Behaviors – Alternate Caregiver Rated. Due to the preliminary finding that prenatal maternal distress significantly predicted externalizing behaviors as rated by an alternate caregiver, we were able to next evaluate whether child cortisol reactivity mediated this relationship. We created a new model in the context of SEM that included cortisol reactivity, and we controlled for the effects of baseline cortisol

measurements on the dependent variable (see Figure 3). Because criteria for mediation mandates that the predictor variable is significantly associated with the mediator and that the mediator is significantly associated with the outcome variable (Rose, Holmbeck, Coakley, & Franks, 2004), these associations were tested prior to adding a direct path between the predictor and outcome variable of the model. The structural model testing these two associations was of adequate fit (χ^2 (*df* = 13) = 19.36, *p* = .11, CFI = .98, RMSEA = .08), such that prenatal maternal distress significantly predicted child cortisol reactivity (β = .18, *p* = .04) and child cortisol reactivity significantly predicted externalizing behavior (β = .38, *p* = .05). These significant regression weights are shown in Figure 3.

Rose et al. (2004) additionally states that in order for a variable to be recognized as a mediator, the relationship between the predictor (i.e. prenatal maternal distress) and outcome (i.e. externalizing behavior) variables must be diminished when the effects of the mediator variable are controlled for. In the context of SEM, this means that the mediator variable must be added to the model that tests the association between the predictor and outcome variable. Thus, a direct path from the predictor to outcome variable was added to the model that originally tested the associations between the mediator and both the predictor and outcome variables (described above). This model achieved adequate fit (χ^2 (*df* = 12) = 15.68, *p* = .21, CFI = .99, RMSEA = .07) and yielded a non-significant association between prenatal maternal distress and externalizing problems (β = .25, *p* = .06) once cortisol reactivity was controlled (see Figure 3).

Attention and Aggressive Problems – Alternate Caregiver Rated. Because SEM indicated that prenatal maternal distress predicted externalizing behavior problems in childhood (as rated by an alternate caregiver), we further examined whether prenatal maternal distress predicted two specific types of externalizing behaviors: (a) attention problems, and (b) aggression problems. Structural models were created to test the main effect of prenatal maternal distress on both of

these outcome variables, and both models achieved good fit (attention: χ^2 (df = 5) = .49, p = .99, CFI = 1.00, RMSEA = .00; aggression: χ^2 (df = 5) = 3.37, p = .64, CFI = 1.00, RMSEA = .00). Significant main effects of prenatal distress were found for both structural models (attention: β = .28, p = .04; aggression: β = .32, p = .02).

Impact of Child Stress Reactivity on Attention Problems – Alternate Caregiver Rated. We next analyzed the potential mediating role of child cortisol reactivity in the relationship between maternal prenatal distress and child attention problems. The mediational model achieved adequate fit (χ^2 (df = 12) = 16.19, p = .18, CFI = .98, RMSEA = .07), however, cortisol reactivity did not mediate the relationship between prenatal maternal distress and attention problems. As shown in Figure 4, the original association between the predictor and outcome variable (β = .32, p = .02) remained significant after controlling for child cortisol reactivity (β = .31, p = .03), and the association between cortisol reactivity and attention problems was not significant (β = .02, p = .91).

Impact of Child Stress Reactivity on Aggressive Problems – Alternate Caregiver Rated. In order to evaluate the potentially mediating effect of child cortisol reactivity in the association between prenatal maternal distress and aggressive behavior (as rated by the alternate caregiver), we first created a structural model that measured the relationship between the potential mediator and the independent and dependent variables. This model achieved an adequate fit for the data (χ^2 (df = 13) = 20.19, p = .09, CFI = .97, RMSEA = .09). Prenatal maternal distress significantly predicted child cortisol reactivity (β = .18, p = .04), and child cortisol reactivity significantly predicted aggression problems (β = .45, p = .02). These associations are shown in Figure 5.

After determining that the potential mediator was significantly associated with both the predictor variable (i.e. prenatal maternal distress) and the outcome variable (i.e. aggressive behavior), we added a direct path from the predictor variable to the outcome variable in order to

determine if controls for child cortisol reactivity would result in a diminished significance of the effect of maternal prenatal distress on child aggression. The structural model maintained adequate fit after adding this path (χ^2 (df = 12) = 16.50, p = .17, CFI = .98, RMSEA = .07). It was found that when controlling for child cortisol reactivity in the structural model, the significant association between prenatal maternal distress and aggression problems disappeared (β = .25, p = .06). It was therefore concluded that the relationship between prenatal maternal distress and aggression problems was mediated by cortisol reactivity of the child (see Figure 5).

Interactions

Eight individual interaction hypotheses were tested during data analyses and these statistics are shown in Table 5 and Table 6. Two different variables were used to reflect the potentially moderating effect of the LES, and this was done because the two variables capture different aspects of the LES. These variables were (a) total number of life events in the past six months (see Table 5), and (b) total rating of life events in the past six months (see Table 6).

The interaction effect of each of these two moderators and child cortisol reactivity was tested with four different outcome variables: (a) maternal ratings of externalizing behaviors, (b) maternal ratings of internalizing behaviors, (c) alternate caregiver ratings of externalizing behaviors, and (d) alternate caregiver ratings of internalizing behaviors. Baseline cortisol, illness on the day of the visit, and recent tooth loss were added as covariates for all regressions. Maternal BDI collected on the day of CBCL completion was added as a covariate when testing maternal ratings of externalizing and internalizing behaviors.

As indicated in Tables 5 and 6, two significant interactions were found. First, it was found that child cortisol reactivity and maternal rating of recent life events in the past six months interacted to significantly predict externalizing behavioral problems as indicated by alternate caregiver CBCL scores (β = -.32, t = -2.15, p = .04). In order to assess the direction of this

interaction, the sample was then split into two groups reflecting those children with either a positive or negative total rating of maternal life events in the past six months. A significant main effect was found for the group exposed to maternally rated negative life events ($\beta = .70$, $t = -2.48$, $p = .02$), such that cortisol reactivity significantly and positively predicted externalizing behavior. In contrast, in the children whose mothers did not have a negative LES rating, cortisol reactivity did not significantly predict externalizing behavior problems ($\beta = .33$, $t = 1.18$, $p = .25$).

A second significant interaction was found during data analyses, such that the total number of life events the mother experienced in the past six months moderated the relationship between child cortisol reactivity and internalizing behavior problems as indicated by the mother's CBCL ratings of child behavior ($\beta = .29$, $t = 2.33$, $p = .02$). Number of life events that the mother experienced – and therefore that the child was exposed to – during the past six months was then split into two groups of low and high in order to assess the nature of this interaction. Experiences of zero or one major life event in the past six months constituted the "low" group, and experience of more than two life events in the past six months constituted the "high" group.

Main effects were then tested for each group and neither regression showed a significant main effect between cortisol reactivity and child internalizing problems ("low" group: $\beta = -.63$, $t = -2.50$, $p = .02$; "high" group: $\beta = .26$, $t = 1.18$, $p = .25$). The pattern of results suggested a negative (significant) relationship between cortisol reactivity and internalizing problems for children exposed to a low number of stressors, and a positive (non significant) relationship between cortisol reactivity and internalizing problems for children exposed to a higher number of recent life stressors.

Discussion

The aim of the current study was to further investigate the risk pathway associated with prenatal maternal distress exposure and behavioral problems in childhood, while accounting for the mediating effect of cortisol reactivity and the moderating effect of the postnatal environment. The main hypothesis, that prenatal maternal distress exposure would predict child behavioral problems during the preschool years, was partially supported. In particular, prenatal maternal distress predicted externalizing problems in childhood (as rated by an alternate caregiver), and was not predictive of internalizing problems in childhood. These findings are consistent with the literature showing increased rates of externalizing behavioral problems such as attention, hyperactivity, and aggressive tendencies in children prenatally exposed to maternal distress (Monk, 2001). It should be mentioned that non-significant findings linking prenatal maternal distress with internalizing problems in childhood might be due to how these behavioral tendencies do not develop until the later years (Mash & Wolfe, 2010).

Research on this topic throughout the past several decades has investigated the role of the HPA axis as the mechanism linking the relationship between prenatal maternal distress and behavioral problems in childhood. Moreover, the literature has concluded that a reliable representation of the HPA axis and its functional regulatory capacities can be quantified by measuring the change in one's cortisol levels from a baseline to post-stressor situation. Sondeijker et al. (2008) clarified that reactivity is a better picture of HPA axis regulation (or dysregulation) than baseline in their study revealing consistent discrepancy in the findings regarding predictors and outcomes associated with this measure of cortisol. Thus, the current study looked at cortisol reactivity as a mediator between prenatal maternal distress and child behavioral problems. Structural equation modeling revealed that cortisol reactivity significantly

mediated the relationship between prenatal maternal distress and externalizing behavior problems in children as rated by an alternate caregiver.

The current study then further examined the relationship between prenatal distress and two particular subtypes of externalizing behavior: attention and aggression. Findings suggested that prenatal maternal distress significantly predicted both attention and aggression, however aggression was the only behavioral subtype that was mediated by cortisol reactivity. The literature investigating the relationship between dysregulation of the HPA axis and aggressive problems has produced mixed results, with some studies showing an inverse relationship between cortisol reactivity and aggression, and some showing that cortisol reactivity positively predicts aggressive problems in childhood (Lopez-Duran, Olson, Hajal, Felt, & Vazquez, 2009). These mixed findings in the literature may be a result of differing lab paradigms and variability in measurement. The results of the current study indicate that higher levels of cortisol reactivity are associated with aggressive problems in childhood. Lopez-Duran and colleagues (2009) recommend that future research investigate the different types of aggression as discrete categories, as it has been found that higher levels of cortisol reactivity are associated with *reactive* aggression (e.g. hostile attribution bias), and not *proactive* aggression (e.g. goals of dominance and revenge) (Mash & Wolfe, 2010). Although our study did not distinguish between proactive and reactive aggression, it is plausible that most preschool aggressive behaviors are reactive in nature, as children in this developmental stage are still working to regulate and control their emotional responses.

The current study also examined the moderational effect of recent maternal stress on the relationship between child cortisol reactivity and internalizing and externalizing behaviors. The findings noted in this study are consistent with the diathesis-stress model (Belsky & Pluess, 2009), which states that "some individuals, due to a "vulnerability" in their make-up…are

disproportionately or even exclusively likely to be affected adversely by an environmental stressor" (p. 885). Specifically, we found that maternal ratings of life events interacted with cortisol reactivity to predict externalizing problems as suggested by alternate caregiver ratings on the CBCL. A positive relationship was found between cortisol reactivity and externalizing problems in childhood in the group that was exposed to negatively rated maternal events, whereas there was no significant association between cortisol reactivity and externalizing behavior in the group whose mothers gave a positive rating to recent life events. This finding demonstrates support for the diathesis-stress model, such that those who maintain the vulnerability of HPA axis dysregulation (e.g. high cortisol reactivity) will show greater externalizing problems in childhood if exposed to higher levels of maternal distress.

We also noted a moderating effect of the total number of life experiences the mother reported in the past six months on the relationship between cortisol reactivity and internalizing behavior according to mother rated CBCL scores. When split into two groups of high and low exposure to maternal stressful life events, it was found that children exposed to a low number of stressors showed a negative (non-significant) relationship between cortisol reactivity and internalizing problems, whereas children exposed to a high number of stressors showed the opposite (and predicted) direction of relationship. This finding also lends support for the diathesis-stress model because children exposed to a higher number of maternal life events showed a positive association between cortisol reactivity and internalizing problems.

Limitations

There were several limitations of the current study. The first limitation was the relatively small sample size (n = 74). Although many significant findings were revealed during data analyses, and the primary hypotheses were supported, the small sample size must be kept in mind. For the most reliable results of structural equation modeling, Kline (2011) suggests that

studies should use sample sizes of 200 participants or greater. Thus, the current findings must be conceptualized in regard to the suggestion that structural equation modeling be performed with a much larger sample. Moreover, because the current study recruited mothers who sought mental health care during pregnancy, there may be an over-representation of women with higher levels of distress during pregnancy and the postnatal period in our sample.

Additionally, the current study quantified stress reactivity by calculating the difference in cortisol levels between the baseline measure (after entering the lab) and the post-stressor measure. Although many potential confounds were assessed during preliminary analyses, there may still be alternative factors that could have confounded the stress reactivity measurement. For instance, it is possible that the anxiety and stress from entering a new environment (e.g. the lab), the child's "baseline" cortisol measurement did not correctly reflect his or her true cortisol baseline.

Finally, the current study did not account for predictive genetic effects on the behavioral outcomes of this high-risk sample. A handful of studies in the literature have begun to examine the role of *both* the prenatal environment and specific genes in the risk pathway for child psychopathology and these studies can yield great insight into what subgroups of children are at particular risk for child behavioral problems (Belsky & Pluess, 2009).

Strengths

A major strength of the current study is the construction of the prenatal maternal distress variable used in data analyses. This variable incorporates maternal symptoms of depression, stress, and anxiety because these symptoms tend to be co-morbid during pregnancy (Brand, 2011). A latent variable was created that combined the total "area under the curve" of all measures, and therefore a total sum of prenatal maternal distress exposure across all of gestation was used in analyses. In addition to measuring different symptoms of maternal distress, the

components of the latent variable came from a variety of assessment techniques, such that some were self-reports and others were monitored by a clinician. By using a combination of prospective measures that were completed by different sources, the latent variable in the current study was able to more validly measure the extent to which the fetus was prenatally exposed to distress.

Moreover, the current study accounted for the potential bias that is often seen when mothers complete questionnaires that measure attributes about their child. As shown by the strong positive relationship between maternal BDI score and maternal CBCL scores, a mother's current symptomology has the potential to influence how she assesses her child on standard questionnaires. The current study, therefore, also collected CBCL questionnaires from an *alternate* caregiver, such as a father, grandmother, or nanny. As the results show, alternate caregiver responses on the CBCL yielded significant findings that are consistent with the literature, whereas maternal responses did not.

Another strength of this study was that it measured different categories and subcategories of child behavior. By distinguishing between internalizing and externalizing behavior, the present study was able to make more specific conclusions regarding the relationship between prenatal maternal distress exposure and behavioral outcomes for the sample. After finding that prenatal maternal distress significantly predicted externalizing behavior problems (as rated by an alternate caregiver), the current study further distinguished between attention and aggressive problems. As discussed previously, these findings illustrate the importance of measuring specific behavioral outcomes during childhood in order to fully understand the developmental pathways of risk from prenatal distress to child outcomes.

Implications

The primary finding of the study is that prenatal maternal distress predicts child externalizing behavior problems. Although somewhat unfeasible, it should be mentioned that one potential way to prevent child behavior problems would be to minimize the distress that mothers experience during pregnancy. The association between prenatal maternal distress and externalizing behavioral outcomes such as inattention and aggression has been replicated in the literature, and the current study adds to the literature by accounting for cortisol reactivity as a mediator of this model. Interestingly, this study also shows that when analyzing behavioral outcomes more specifically, HPA axis dysregulation may mediate the relationship between prenatal maternal distress and some externalizing behaviors but not others. Research regarding the components of the risk pathway leading to different externalizing behaviors in childhood can lead to a better understanding of what psychopathologies can be expected (and hopefully prevented) in children at high-risk for mental health issues later in life.

McMahon (1994) describes externalizing behaviors as "acting out" behaviors, and asserts that the two main disorders that can result from externalizing problems in childhood are conduct disorder and ADHD. Much research supports the notion that externalizing problems in childhood are highly predictive of externalizing problems in adulthood, and that individuals who display the highest levels of externalizing problems in childhood are at the greatest risk for psychopathological problems in adulthood (Wolfe & Mash, 2010). Therefore, it is important to study these behaviors in early childhood and gain a better understanding of what variables predict these outcomes due to the undesirable prognosis that is often associated with children who demonstrate externalizing problems at very early ages (McMahon, 1994).

It is not surprising that dysregulation of the HPA axis significantly mediated the relationship between prenatal maternal distress and externalizing problems in the current sample,

given the regulatory capacity reflected by this system and the regularity issues associated with externalizing behaviors. What is most interesting about the findings of the current study is that when we investigated the impact of cortisol reactivity on the relationship between different subtypes of externalizing behaviors, and findings showed that HPA axis dysregulation only predicted aggressive behavior problems and not attention problems. The mediating role of HPA axis dysregulation on the relationship between prenatal maternal distress exposure and aggressive problems in childhood may be due to the lack of self-regulation that is associated with HPA axis reactivity (Bauer, Quas, and Boyce, 2002). A study conducted by Granger, Weisz, and Kauneckis (1994; as cited in Bauer et al., 2002) found that children with higher cortisol reactivity were more likely to feel that they have little personal control in their lives. These children may act out in an aggressive manner in an attempt to gain control over their environment. They may see the world as more hostile and unpredictable than it is, which is consistent with cognitive theories of aggression (Dodge & Pettit, 2003).

Recently the literature has begun to differentiate between types of childhood aggression, and these distinctions can be useful for both intervention and prevention programs. Research demonstrates that children who have high cortisol reactivity demonstrate tendencies associated with *reactive* aggression (Lopez-Duran et al., 2009), which is a type of regression associated with attributing hostile intent to others. These findings suggest the need for more research regarding the associations between prenatal maternal distress and specific types of aggression.

Results of the current study also indicate that recent maternal stressful life events may moderate the relationship between HPA axis dysregulation and externalizing and internalizing problems in childhood. This is a promising finding that supports the diathesis-stress model, such that those who maintain the vulnerability of HPA axis dysregulation will experience greater levels of behavior problems only when concurrently exposed to a stressful postnatal

environment. This interaction creates a promising picture of the strong influence that the postnatal environment can have on children who are predisposed to the development of psychopathology. Furthermore, in correspondence with Belsky and Pluess's (2009) argument of "differential plasticity", it may be possible that children who are predisposed to developing psychopathology may also be the most susceptible to the protective effects of the environment.

Future Research Directions

Further research regarding the applicability of the fetal programming hypothesis to psychological and psychiatric outcomes is also of prime importance. Ellison proposes that public health interventions are possible if research begins to examine the consequences that can result from a 'mismatch' of the fetal and post-natal environment. For instance, experimental paradigms that 'match' and 'mismatch' prenatal and postnatal environments can yield more conclusive claims of the fetal programming hypothesis and deepen our understanding of how the fetus undergoes adaptation during prenatal development (Ellison, 2010). By developing a greater understanding these developmental mechanisms, strategies can be developed in order to avoid negative consequences associated with prenatal maternal distress exposure (Ellison, 2010). Furthermore, with Belsky and Pluess's (2009) concept of "differential susceptibilities" in mind, it may be more proactive for future research to study environmental factors that will have a *protective* effect on high-risk groups, for these are the variables that are most important to intervention and prevention programs.

Conclusions

Ample research conducted over the past several decades has clarified the importance of the prenatal environment for optimal fetal and postnatal development. The current study corroborates the importance of the fetal environment by demonstrating that children who are prenatally exposed to high levels of maternal distress are at great risk for the development of

externalizing behavioral problems during the preschool years. It was also found that child cortisol reactivity mediated the relationship between prenatal maternal distress and externalizing problems (specifically, aggression). Additional findings highlight the importance of a positive (i.e., less stressful) postnatal environment in minimizing negative behavioral outcomes associated with HPA axis dysregulation.

By analyzing the mediating effect of cortisol reactivity and the moderating effect of current maternal stress, this study added to the literature by investigating external variables that are involved in the risk pathway leading to child psychopathology. Overall, findings from the present study indicate a need for clinicians to be cognizant of maternal distress levels throughout pregnancy, for fetal exposure to maternal distress may lead to HPA axis dysregulation and resulting externalizing problems in childhood. Future research investigating the most effective therapies during both the prenatal and postnatal years is needed in order to minimize the potential harm that maternal distress can have on children at high-risk for the development of psychopathology.

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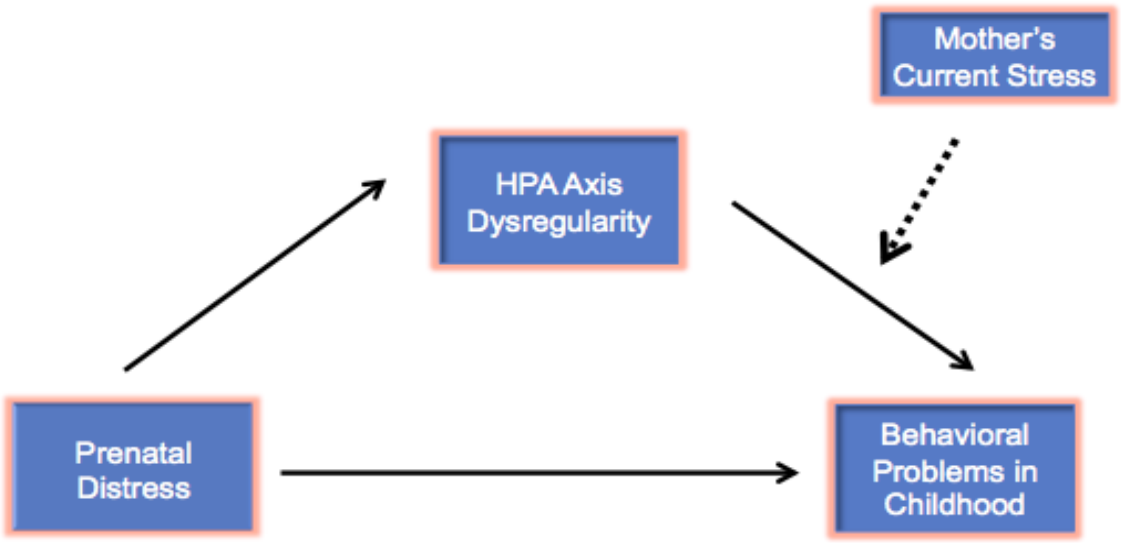
Figure 1.

Study model



Notes: This figure illustrates a conceptual model of the study's hypotheses.

Figure 2.

Association between prenatal maternal distress and externalizing problems (as rated by an alternate caregiver)

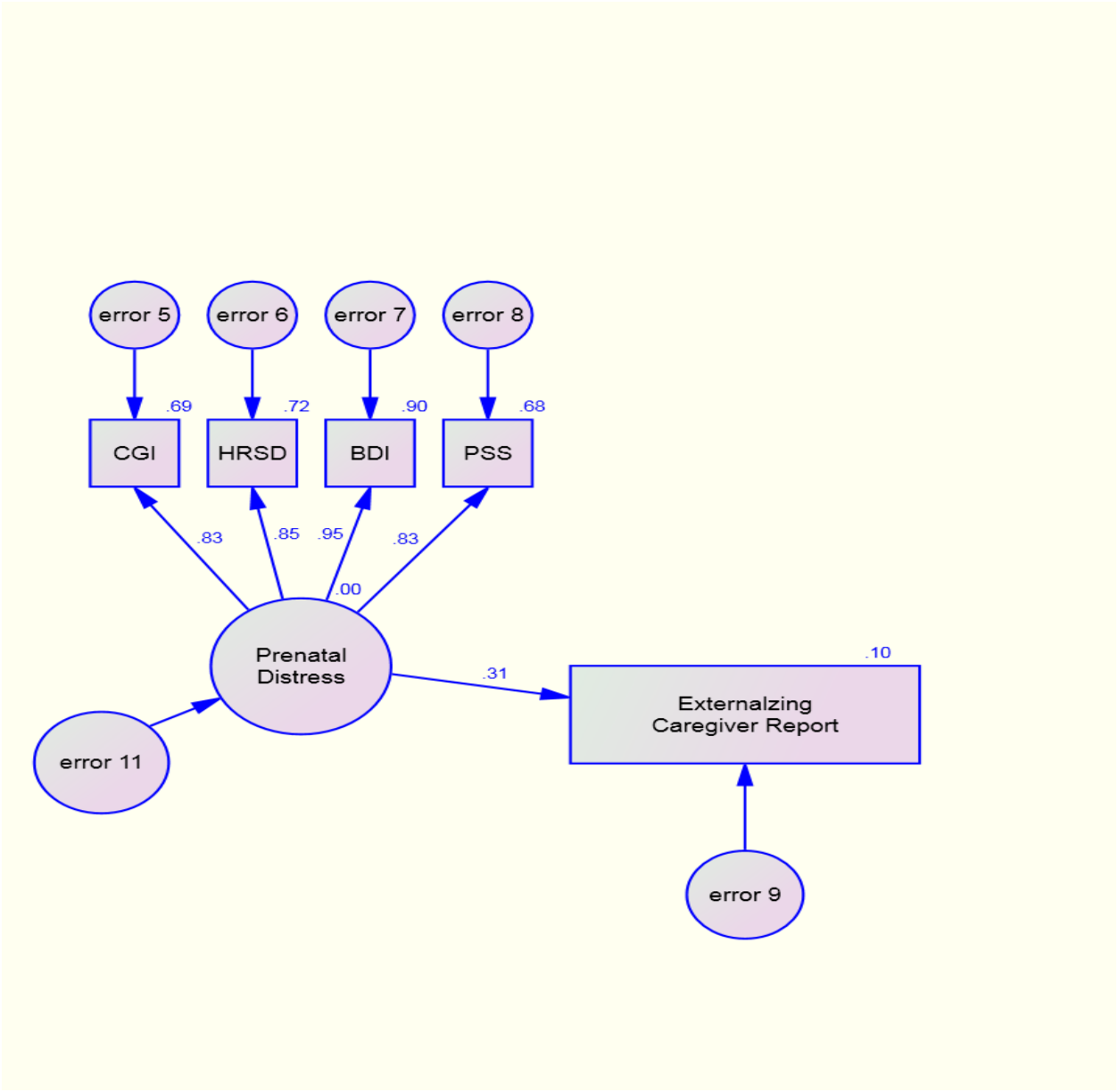
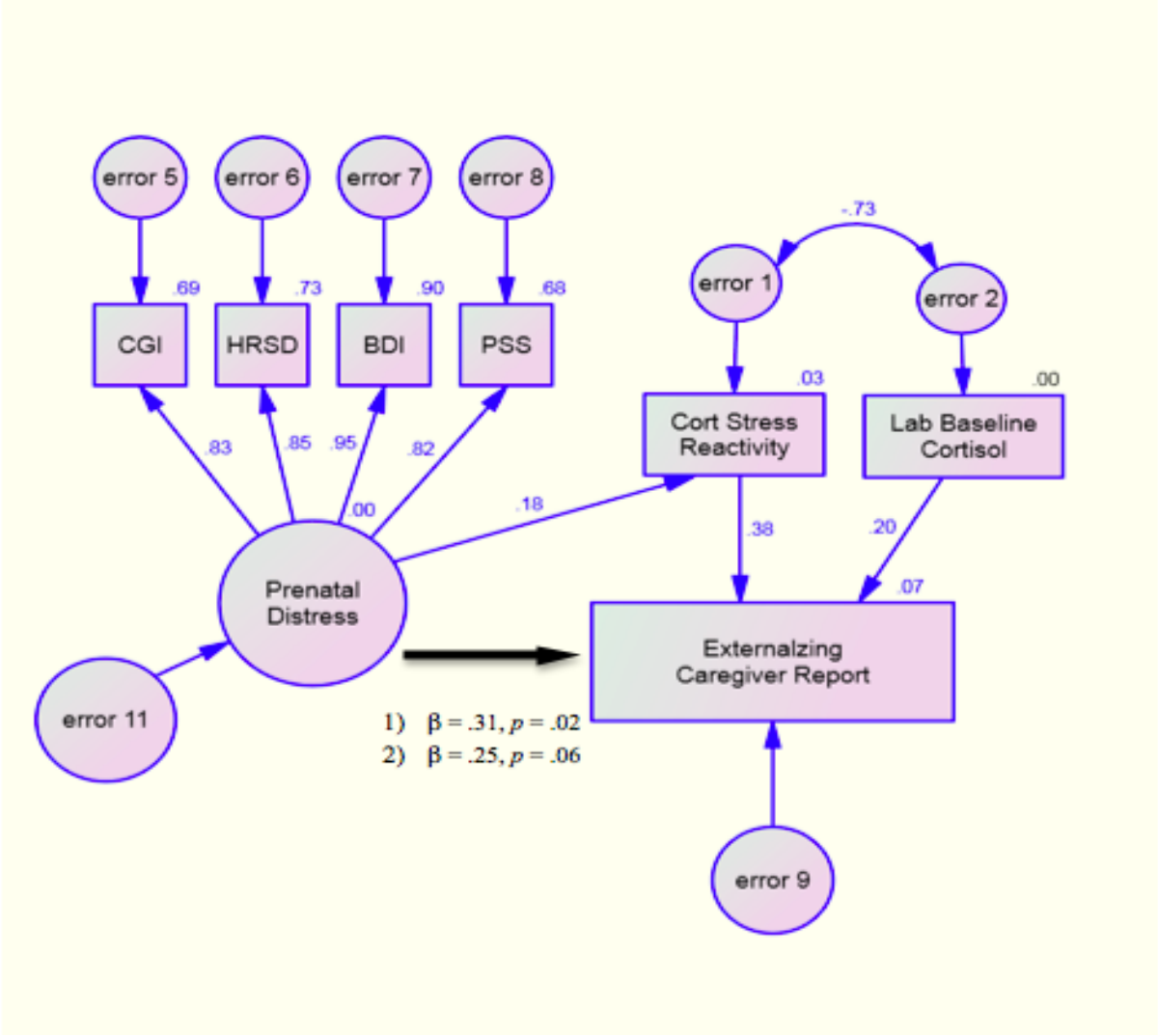


Figure 3.

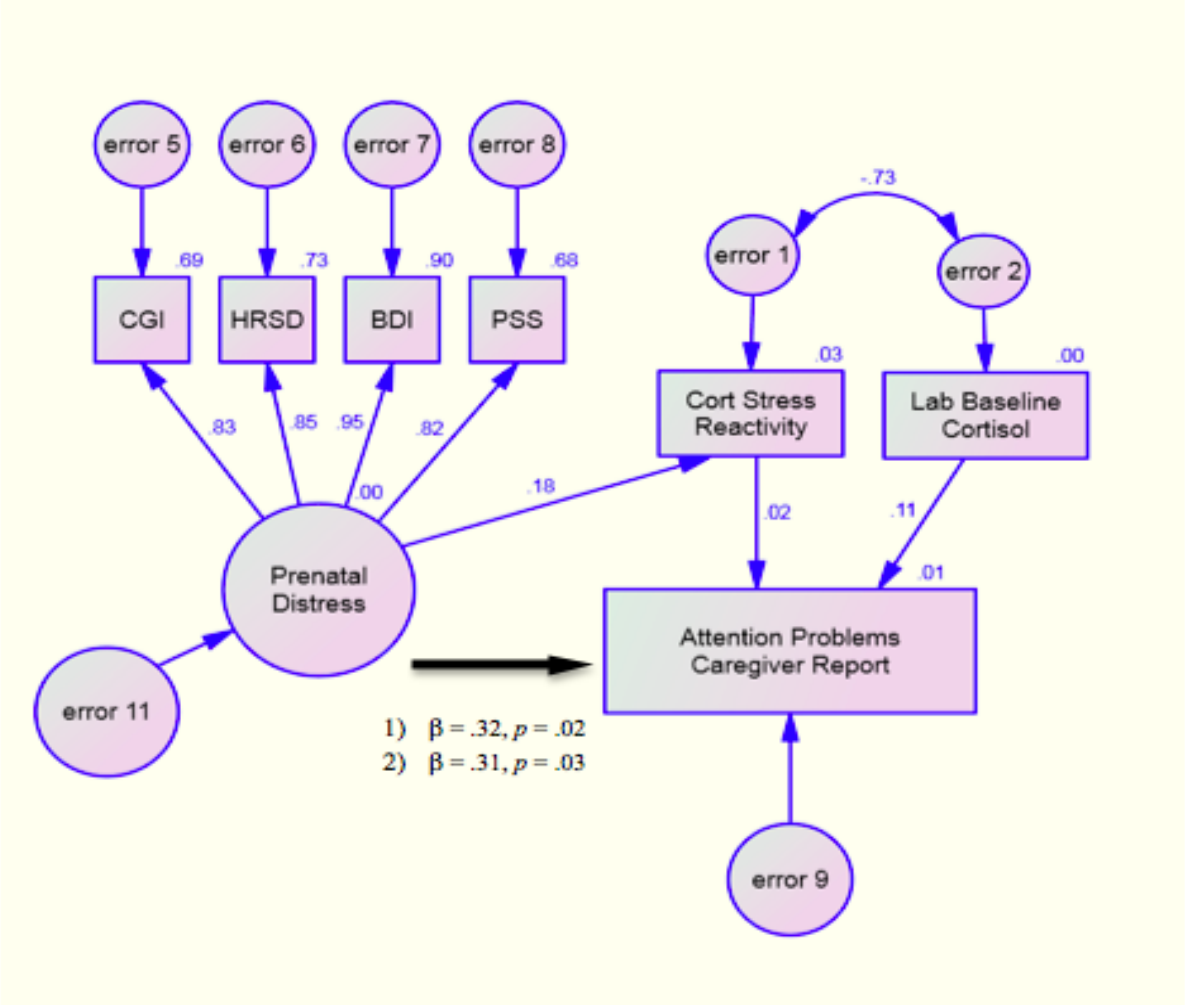
Impact of child stress reactivity on the association between prenatal maternal distress and externalizing child behavior (as rated by an alternate caregiver)



Notes: The first (1) set of statistics presented above under the path between prenatal distress and externalizing child behavior reports illustrate the significant association between the two variables *before* controlling for the mediating effect of child cortisol reactivity. The second (2) set of statistics presented demonstrate the non-significant association between these variables after accounting for child cortisol reactivity in the structural model.

Figure 4.

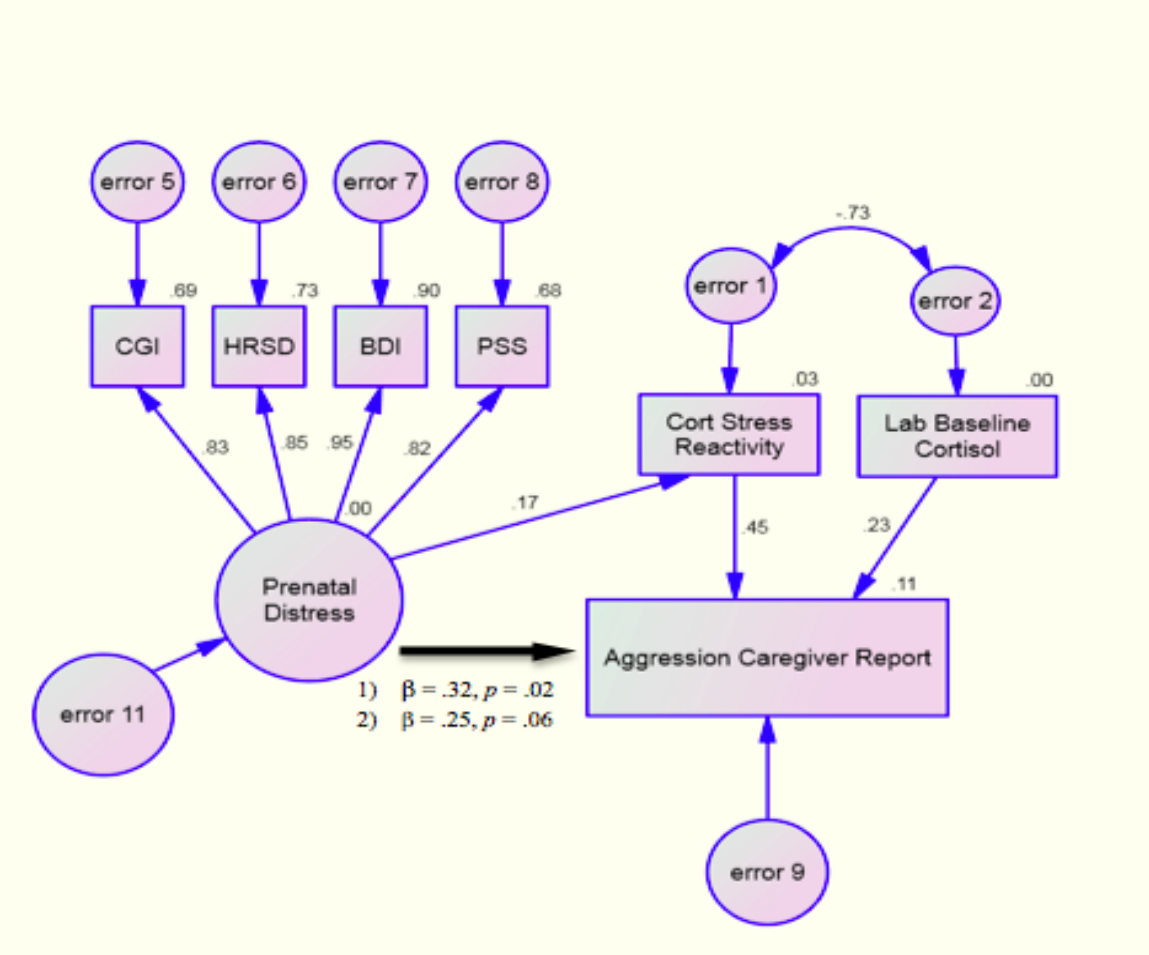
Impact of child stress reactivity on the association between prenatal maternal distress and attention problems of the child (as rated by an alternate caregiver)



Notes: The figure above illustrates that child stress reactivity does not mediate the relationship between prenatal maternal distress and attention problems as indicated by an alternate caregiver report. The second (2) set of statistics showed that this association remained significant after controlling for child cortisol reactivity. Mediation of child stress reactivity was not plausible because child stress reactivity did not predict attention problems, as shown by the low regression weight (β = .02).

Figure 5.

Impact of child stress reactivity on the association between prenatal maternal distress and aggression problems of the child (as rated by an alternate caregiver)



Notes: The first (1) set of statistics presented above under the path between prenatal distress and aggressive child behavior reports illustrate the significant association between the two variables *before* controlling for the mediating effect of child cortisol reactivity. The second (2) set of statistics presented above demonstrate that after accounting for child cortisol reactivity in the structural model, the association between prenatal distress and aggressive child behavior disappeared.

Table 1

Prenatal Maternal Distress Components: Explanation of Questionnaires

Measure	Description
Hamilton Rating Scale for Depression (HRSD)	A semi-structured interview conducted by a research assistant that contains 17 items. Participants rate symptoms of depression such as guilt and feelings of worthlessness and total scores range from 0-81. Higher scores represent higher levels of depression.
Clinical Global Index (CGI)	A questionnaire completed by the patient's clinician yielding a global rating of the severity of the patient's symptoms of the certain disorder that he or she has.
Beck Depression Inventory (BDI)	A self-report measure consisting of 21 items. Patients rate symptoms that are divided by categories from 0-3, and a summed score yields the patient's level of depression. A score of 13 is indicative of depression, and as scores increase, so does the severity of the depression. The BDI measures depression symptoms of the past two weeks.
Perceived Stress Scale (PSS)	A self-report measure consisting of 14 items that measure the patient's feelings about stressors in his or her life. Answers range from 0-4, with higher ratings indicating that the certain stressor occurs more often.

Notes: The information regarding these four questionnaires was taken from Sarah Brand's (2011) dissertation, *Maternal psychosocial distress during pregnancy and child behavior outcomes* due to similarity of variable construction between the dissertation and the current thesis.

Table 2

Inter-correlation between measures of maternal distress during pregnancy (AUC)

		CGI	BDI	HRSD	PSS
CGI	Pearson Correlation	--	.793**	.690**	.681**
	Sig. (2-tailed)		.000	.000	.000
BDI	Pearson Correlation	--	--	.800**	.783**
	Sig. (2-tailed)			.000	.000
HRSD	Pearson Correlation	--	--	--	.712**
	Sig. (2-tailed)				.000
PSS	Pearson Correlation	--	--	--	--
	Sig. (2-tailed)				

**. Correlation is significant at the 0.01 level (2-tailed).

Notes: This table demonstrates the high and positive correlation between the four independent prenatal distress measures. The high correlations between all measures indicate that construction of a latent variable is appropriate.

Table 3.

Testing for Potential Confounds Associated with Child Behavioral Measures

		Child Sex	Child Age	Maternal BDI on Day of Visit	Prenatal Psychiatric Illness Exposure	Mother's Age	Mother's Education
Alternate CBCL: Internalizing	Pearson Correlation	.051	.024	.215	.054	-.160	-.077
	Sig. (2-tailed)	.705	.859	.109	.694	.238	.572
Alternate CBCL: Externalizing	Pearson Correlation	.040	-.228	.196	.121	-.166	.017
	Sig. (2-tailed)	.768	.088	.145	.374	.221	.902
Alternate CBCL: Attention	Pearson Correlation	-.086	-.235	.160	.005	-.178	.085
	Sig. (2-tailed)	.527	.078	.234	.971	.190	.532
Alternate CBCL: Aggressive	Pearson Correlation	.048	-.213	.197	.166	-.156	-.013
	Sig. (2-tailed)	.721	.112	.142	.221	.251	.921
Maternal CBCL: Internalizing	Pearson Correlation	.062	.134	.243**	.206	.157	.078
	Sig. (2-tailed)	.600	.258	.038	.084	.188	.513
Maternal CBCL: Externalizing	Pearson Correlation	-.029	.002	.238**	.129	.069	.095
	Sig. (2-tailed)	.806	.986	.043	.282	.567	.429
Maternal CBCL: Attention	Pearson Correlation	-.047	-.002	.181	.142	-.121	.184
	Sig. (2-tailed)	.695	.985	.126	.236	.311	.123
Maternal CBCL: Aggressive	Pearson Correlation	-.021	.003	.231**	.114	.116	.060
	Sig. (2-tailed)	.857	.979	.049	.345	.331	.614

**. Correlation is significant at the 0.01 level (2-tailed).

Notes: Maternal BDI was the only significant confound associated with behavioral outcome measures.

Table 4

Descriptive Statistics of Independent, Dependent, Moderating, and Mediating Variables

	N	Minimum	Maximum	Mean	Std. Deviation
CGI (AUC)	71	40	182	74.24	33.115
BDI (AUC)	74	29.80	1464.58	440.9284	347.36033
HRSD (AUC)	72	130.93	907.58	439.3640	180.04361
PSS (AUC)	74	379.63	1828.49	1017.6501	336.49304
Attention (Alt)	57	0	6	1.84	1.688
Aggressive (Alt)	57	0	26	8.12	6.585
Internal (Alt)	57	0	32	6.46	5.985
External (Alt)	57	0	31	10.18	7.700
Attention (mother)	73	0	8	1.68	1.739
Aggressive (mother)	73	0	24	8.84	6.160
Internal (mother)	73	0	29	7.99	5.095
External (mother)	73	0	30	10.52	7.297
LES Total Events	74	0	13	3.47	2.934
LES Total Rating	74	-20	17	-1.47	6.089
Cortisol Baseline	71	.01	.80	.1607	.16659
Cortisol Reactivity	71	-.64	.73	-.0142	.19686

Table 5.

Regression Analysis Statistics for Interactional Effect of LES Total Events x Child Stress Reactivity on Behavioral Problems in Childhood

	F change	β	*t*	*p* value
Alternate CBCL: Internalizing	.89	.14	.94	.35
Alternate CBCL: Externalizing	.04	.03	.20	.84
Maternal CBCL: Internalizing	5.41	.29	2.33	.02*
Maternal CBCL: Externalizing	.79	.12	.89	.38

*. Correlation is significant at the 0.05 level (2-tailed).

Table 6.

Regression Analysis Statistics for Interactional Effect of LES Total Rating x Child Stress Reactivity on Behavioral Problems in Childhood

	F change	β	*t*	*p* value
Alternate CBCL: Internalizing	.48	-.11	-.69	.49
Alternate CBCL: Externalizing	4.64	-.32	-2.15	.04*
Maternal CBCL: Internalizing	.01	.01	.10	.92
Maternal CBCL: Externalizing	.15	-.06	-.39	.70

*. Correlation is significant at the 0.05 level (2-tailed).