

THE ROLE OF THE AUTONOMIC NERVOUS SYSTEM IN EARLY LIFE STRESS
AND INTERNALIZING SYMPTOMS

A Senior Honors Thesis

Presented to

The Faculty of the Department

of Psychology

University of Houston

In Partial Fulfillment

Of the Requirements for the Degree of

Bachelor of Science

By

Catherine A. Ramos

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ABSTRACT

Early life stress exposure has significant deleterious psychological effects that can put youth at risk for internalizing problems such as anxiety and depression. In this study, we examined how early life stress shapes physiological correlates of emotional and stress regulation, as predictors of increased depression and anxiety risk in early childhood. Data were drawn from a preliminary sample of preschool-aged children, between 4 to 6 years of age. In a laboratory session, parents reported on several domains of early life stressors: family income, lack of family resources, lack of family support, and exposure to domestic violence. Children participated in an emotional induction task, where autonomic nervous system (ANS) variability was assessed. Parents also reported on child internalizing symptoms. Results indicated that among risk factors, lack of family resources and exposure to domestic violence, were associated with heart rate (HR) reactivity to sad and scary emotional stimuli. Results also supported links between RSA and HR reactivity to sad emotional stimuli and risk for depression symptoms in early childhood. In summary, findings suggest that early life stress may increase physiological reactivity to negative stimuli, and this may be a predictor internalizing symptoms risk. This study emphasizes the importance of studying these constructs in highly diverse samples to further the understanding of ANS activity with emotion dysregulation.

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THE ROLE OF THE AUTONOMIC NERVOUS SYSTEM IN EARLY LIFE STRESS AND INTERNALIZING SYMPTOMS

Accumulating evidence suggests that exposure to early life stress can increase the risk for internalizing problems such as anxiety or depression symptoms (Cohen, Brown & Smailes, 2001; Green et al., 2010; McLaughlin et al., 2012; Scott, McLaughlin, Smith, & Ellis, 2012). Early life risk factors also have long term consequences. Longitudinal studies have shown that early life stressors, such as parental depression, early childhood internalizing symptoms, childhood adversity, and low socioeconomic status (SES), predicted later development of depression in adulthood (Gilman, Kawachi, Fitzmaurice, & Buka, 1995; Jaffee et al., 2002; Johnson, Cohen, Dohrenwend, Link, & Brook, 1999; Kessler & Magee, 1993; Reinherz, Giaconia, Hauf, Wasserman, & Paradis, 2000; Sadowski, Ugarte, Kolvin, Kaplan, & Barnes, 1999). However, not all children exposed to stressful environments develop psychopathology.

Burgeoning work suggests that early life stress also affects the development of underlying physiological systems, such as the autonomic nervous system (ANS), which supports stress response and relates to the capacity for emotion regulation. Alterations in these systems may point to mechanisms underlying risk for the development of internalizing symptoms. Less is known concerning which early life stressors impact ANS variability, and whether these pathways help predict risk for depression and anxiety in early childhood. Therefore, the goal of this study was to examine associations between autonomic reactivity and risk for internalizing symptoms in a high-risk sample of children who varied in early life stressors.

Autonomic Nervous System

The body has two main central stress response systems, the hypothalamic pituitary adrenal axis, which is slow acting and releases cortisol, and the autonomic nervous system, which is fast acting at preparing the body for stress. The autonomic nervous system is composed of the two branches: the sympathetic (SNS) and parasympathetic nervous system (PNS). The ANS is a complex system that monitors the internal functioning of the body to modulate towards stability regarding shifts from external or internal stressors. The ANS is not just static until provoked by stressors but is constantly monitoring and facilitating necessary adjustments to maintain homeostasis (Porges, 1995). Both systems modulate neural control from the brainstem to inhibit or accelerate activity in specific organs such as the heart, intestines, glands, and other related structures (Porges, 1995).

The sympathetic nervous system (SNS) is responsible for activating the body's physiological arousal during situations of stress in preparation for fight-or-flight, by producing glucose, increasing heart rate, pupil dilation, and other biological processes (Porges, 1995). Influence from the PNS adjusts bodily functions towards features of "rest-and-digest," such as preserving energy through slowing of heart rate and readying the body for proper food digestion, such as adjustments of salivary flow and intestinal regulation (Cannon, 1929; Porges, 1995). The PNS is necessary for modulating recovery from sympathetic arousal from stressors, maintaining homeostasis, and supporting stress activation to challenging situations (Porges, 1995). This is accomplished through the increasing or decreasing of parasympathetic activity which functions as a "brake" on the expression of sympathetic activity on the body. However, the interaction between the two

branches of ANS do not always result in mutual activation and deactivation (Porges, 1995).

In general, there are 3 observed patterns of different SNS & PNS activations which includes reciprocal sympathetic activation, reciprocal parasympathetic activation, and nonreciprocal activation. Reciprocal sympathetic activation is when the sympathetic activates and parasympathetic is inhibited, which allows for adrenal response within the individual through physiological expression such as heart rate increase and pupil dilation. Reciprocal parasympathetic activation is the sympathetic inhibition and parasympathetic activation, which would allow for emotional regulation, attention, and calmness as heart rate is slowed and other basal functioning resumes. Lastly there is nonreciprocal activation, to which the responses from the autonomic nervous system are opposing each other, which has been theorized in having implications of emotional dysregulation due to asynchrony within the ANS (El-Sheikh & Erath, 2011).

There are two types of nonreciprocal activation known as coactivation and coinhibition. Coactivation is the increased sympathetic action and increase parasympathetic action that oppose each other, whereas coinhibition is the decrease in both sympathetic and parasympathetic action in opposition. It is theorized that in episodes of nonreciprocal activation within the ANS, there might be a “null” physiological response in that no change is made within the system due to the opposing activations equaling out (El-Sheikh & Erath, 2011). However, it is not known if one branch of the autonomic nervous system has more influence than the other in regulating these physiological responses, or if an “equaled” response in both branches is maladaptive in stressful situations that require action. A few studies have shown that

co-inhibition is present in children with emotional dysregulation such as display of externalizing symptoms during challenging tasks (Beauchaine, Gatzke-Kopp, & Mead, 2007; El-Sheikh & Erath, 2011).

Heart rate (HR) is an important indicator of combined physiological sympathetic and parasympathetic activity of the autonomic nervous system and has been used as an indicator of dominant sympathetic physiological activity when reacting to challenging or threatening stimuli. There is a typically observed inverse relationship with the indices respiratory sinus arrhythmia (RSA) and HR; when RSA increases, HR decreases due to the parasympathetic activation slowing the heart (Dietrich et al., 2007). Because so many processes influence heart rate, it is not considered a specific index of either the sympathetic or parasympathetic system. Porges (1995) emphasized that measurement and assessment of parasympathetic influence would provide a better understanding on the modulation of both branches of ANS during stress, rather than just sympathetic activity alone.

Respiratory sinus arrhythmia (RSA). Parasympathetic influence is most notably measured through the sensitive heart rate variability index of respiratory sinus arrhythmia (RSA) (Porges, Doussard-Roosevelt, Portales, & Greenspan, 1996). Respiratory sinus arrhythmia (RSA) is a non-invasive indicator of PNS vagal activity that measures the natural variation in heart rate (HR) that occurs with respiration. The PNS acts through the vagal nerve as an inhibitory influence on the heart. Polyvagal theory indicates that vagal input is responsible for respiratory sinus arrhythmia (RSA) on heart rate variability. RSA reflects the body's natural heart rate variations in relation to the respiratory cycle, such that HR is slower during exhalation and faster during inhalation

(Eckberg & Eckberg, 1982). RSA also indexes vagal nerve input, which starts at the brain stem and ends at the heart's pacemaker, which allows for parasympathetic control over the heart's variability and therefore control over bodily resource utilization or conservation. Resting RSA is measured at one timepoint and represents an individual's potential responsiveness to the environment and ability to regulate emotional states. There is substantial literature that consistently associates higher resting RSA with greater abilities to regulate stress responsivity, emotional arousal, and attention (Bornstein & Suess, 2000; Porges, et al., 1996; Stifter & Fox, 1990). Lower resting RSA indicates vagal input at rest and decreased HR variability, and has been implicated with the greater psychophysiological arousal observed in chronic stress activation and anxiety disorders (Friedman & Thayer, 1998; Kawachi, Sparrow, Vokonas, & Weiss, 1995; Yeragani et al., 1993).

RSA reactivity reflects shifts from this baseline variability towards or from stressors. Increased vagal activity acts as a "brake" on the heart which slows down HR and sympathetic activity. This increase in vagal activity is referred to as RSA augmentation, and is an indicator of active regulation from the parasympathetic nervous system. Alternatively, the removal of the vagal "brake" allows the SNS to be activated for "fight-or-flight" functionality (Porges, 1995). This is referred to as RSA withdrawal, indicating less vagal regulation from the parasympathetic nervous system. This allows the body to become physiologically aroused via the sympathetic nervous system to address the apparent stressor to the individual (Graziano & Derefinko, 2013). Polyvagal theory suggests that this decrease in RSA (vagal withdrawal) in response to stressful or challenging conditions indicates successful vagal regulation (Porges, 2006), and has been

shown to be associated with better self-regulation, coping skills, and emotion regulation (Degangi, DiPietro, Greenspan, & Porges, 1991; Gentzler, Santucci, Kovacs, & Fox, 2009).

Associations between early life stress and ANS activity: According to the allostatic load theory, the “wear and tear” on the body due to exposure to constant environmental stressors has been shown to advance the human aging process, decrease cognitive functioning, and increase risk of internalizing symptoms (Lynch, Kaplan, & Shema, 1997; McEwen & McEwen, 2017). The present environmental stressors, whether consciously or unconsciously perceived as threats to the individual, can cause activation of the limbic system brain regions, which further activates the hypothalamus to signal the HPA axis, and then modulate the autonomic nervous system through release of biochemicals and hormones from the pituitary gland and adrenal gland (McEwen & McEwen, 2017). There is evidence that early life stress is associated with disruptions in ANS functioning, a marker of increased allostatic load. Specifically, children exposed to adversity exhibit altered ANS functioning including lower RSA baseline (Skowron et al., 2011) and less RSA suppression, which may increase risk for internalizing symptoms later on.

Internalizing Symptoms and RSA

A recent meta-analysis examined measures of RSA baseline and RSA reactivity with child internalizing and externalizing symptoms and reported that greater RSA withdrawal to a stressor was associated with higher internalizing and externalizing symptoms (Graziano & Derefinko, 2013). Further, Beauchaine (2015) also suggests that excessive RSA withdrawal may confer risk for psychopathology. However, within

clinical or at-risk samples, lower RSA withdrawal was associated with increased psychopathology (Yaroslavsky, Rottenberg, & Kovacs, 2013). This suggests that contextual factors play an important role in the adaptation of physiological reactivity.

For instance, studies have shown associations between marital conflict and children's autonomic reactivity (El-Sheikh, Keiley, Erath, & Dyer, 2013; Obradović, Bush, & Boyce, 2011). Low baseline RSA with low SNS activity or increasing RSA with decreasing SNS activity to challenge were associated with increased internalizing symptoms in the context of families with high marital conflict (El-Sheikh et al., 2013). However, there is inconsistent support for associations between internalizing symptoms and degree of RSA withdrawal or baseline RSA (Hastings et al., 2008a). Hagan, Roubinov, Adler, Boyce, & Bush (2016) observed that children that are more physiologically reactive in adverse environments are more susceptible to mental health issues such as internalizing symptoms. Davis, Suveg, Whitehead, Jones, & Shaffer (2016) showed that RSA withdrawal may be unnecessary and maladaptive for children in lower-risk environments, but necessary for children in higher-risk contexts to be able to cope with adversity.

Indeed, it does seem that the contextual basis of a child's environment has an important influence in how RSA withdrawal and baseline may be advantageous or disadvantageous, but there is inconclusive evidence as to how (Ellis, Boyce, Belsky, Bakermans-Kranenburg, & Van Ijzendoorn, 2011). For example, theoretical models such as Biological Sensitivity to Context refers to early developmental experience calibrating the body's response system. The model indicates that individuals with heightened stress reactivity profiles can occur in both highly adverse and highly protected early

environments (Boyce & Ellis, 2011). The Adaptive Calibration Model (Del Giudice, Ellis, & Shirtcliff, 2011) also suggests that early alterations to the stress response system can influence long-term functioning within the system in terms of both physiology and behavior.

Internalizing Symptoms and SNS. Similar to the relationship between baseline RSA and RSA suppression with internalizing symptoms, there is limited work on the association between the sympathetic nervous system reactivity and internalizing symptoms in contexts of early stress (Graziano & Derefinko, 2013). Higher levels of HR reactivity may signify autonomic over-arousal to challenging tasks. This over-arousal is theorized as the behavioral output of fearfulness and risk-aversion in challenging or risky situations, and a precursor to anxiety symptoms and internalizing problems (Dietrich et al., 2007). Another physiological index that has given insight to SNS output (HR) but is not included in this study is pre-ejection period (PEP); high SNS activity is indexed by shorter PEP intervals, which correlates with a faster heart rate and increased cardiac output, whereas less SNS activity is marked by longer PEP intervals. Though PEP is a more sensitive biomarker of SNS, directionality of ANS output should be considered by observing heart rate reactivity.

Present Study Aims and Hypotheses

Previous work has shown that increased exposure to early life stress is associated with lower RSA baseline (Gray, Theall, Lipschutz, & Drury, 2017) and variability in RSA reactivity. Additionally, higher levels of internalizing symptoms were associated with lower RSA withdrawal to sadness-inducing tasks in children with higher risk exposure (Davis et al., 2016). These findings support growing evidence for altered

parasympathetic regulation in association with early life stress. There has been less focus on whether similar alterations exist in sympathetic branches of the ANS. A goal of this study is to contribute to this current gap in the literature.

Although early life stress has generally been associated with altered ANS function, less is known on which specific early environmental risk factors may be driving these associations. Furthering the understanding in this area has critical implications for early prevention and intervention. To address this second gap in the literature, the present study examines multiple early life risk factors (SES, family resources, family support, family violence) in a sample of high-risk low SES preschool aged children.

In terms of our overarching questions, we hypothesized that increased childhood risk would be associated with lower ANS functioning at baseline and in response to an emotion induction task. We further hypothesized that ANS variability, in parasympathetic and sympathetic branches, would be associated with increased risk for internalizing symptoms. The study aimed to answer two questions:

Question 1: Does early childhood environmental risk, (low family income, lack of family support, reduced parental resources, and increased family violence exposure) correlate with altered parasympathetic (RSA) and sympathetic (HR) nervous system activity during an emotion induction task?

Question 2: To what extent is variability in parasympathetic (RSA) and sympathetic (HR) nervous system activity during an emotional induction task significantly associated with children's risk for internalizing symptoms?

METHODS

Participants

The participants of this study ($N = 31$) were preschool-aged children ($M_{\text{age}} = 5.0$ years, $SD_{\text{age}} = 1.0$) and their primary caregivers. Families were recruited by posting fliers at community events, WIC centers, and Head Start programs in the Houston area that serve low-income families. Both mothers and children were required to be fluent in English due to English-only measures and tasks. Sample demographics are presented in Table 1. All methods in this study were approved by the Institutional Review Board at the University of Houston.

Procedure

Following recruitment, families were invited to the lab for in-person data collection sessions. Mothers provided consent and children provided assent. Mothers completed a series of questionnaires on their socio-economic status (income, family resources), perceptions of family support, exposure to domestic violence, and their child's behavior.

Children participated in a series of tasks that assessed self-regulation, executive functioning, and emotion regulation. Autonomic nervous system functioning was assessed in a series of tasks with parents and children, including an emotion induction video task examined as part of the current study. At the end of the session, parents received a \$25 gift card and the child received a toy as compensation for participation.

Measures

Family income. Family income was assessed as part of a questionnaire of various demographic characteristics. Mothers reported family income from their personal job, their partners' income, income received as support from government assistance programs, and any other outside sources. All reported sources of income were summed for total family income.

Family Resources and Support. Parents reported on perceived family resources using the Family Resource Scale (FRS; Dunst & Leet, 1987) and Family Support Scale (FSS; Dunst, Trivette, & Cross, 1986). The FRS has adequate reliability with alpha's ranging from .72-.84, and construct validity has been established in a large national sample (Van Horn, Bellis, & Snyder, 2001). For the FSS, Cronbach's coefficient alpha was reported to be .75-.77, internal consistency ranged from .35-.80 for subscales and total scores, and concurrent validity was established with parent and family outcomes (Dunst, Jenkins, & Trivette, 1984). The FSS has been utilized as an indicator of perceived social support the parent has when caring for their pre-school aged child (Dunst et al., 1986). The FSS determines the satisfaction of 18 different sources of support and is rated on a 5-point scale. The FRS and FSS were scored using a sum of each scale. The Cronbach's alpha for the present study were 0.93 and 0.96 for the FRS and FSS dimensions, respectively.

Family Violence Exposure. The Conflict Tactics Scale-2 (CTS) was used to measure domestic violence within the household (Straus, Hamby, Boney-McCoy, & Sugarman, 1996). The CTS has demonstrated satisfactory construct validity, test-retest reliability and internal consistency. The CTS is 80-items and measures psychological and physical attacks between domestic partners (Straus et al., 1996). Parents reported the

frequency of each item in the past year and if ever occurred. The CTS was scored to reflect the number of total events that had occurred in the last year between partners.

Child internalizing symptoms. Mothers were asked to report on their child's behaviors which included internalizing symptoms. Child internalizing symptoms was assessed with the Behavior Assessment System for Children, Second Edition (BASC-2; Reynolds & Kamphaus, 2004). Anxiety and Depression subscales were calculated from BASC-2. Adequate levels of internal consistency, test-retest reliability, and validity have been previously established (Reynolds & Kamphaus, 2004).

Autonomic Nervous System Data

Emotion induction paradigm. To assess ANS reactivity, children engaged in an emotion induction task designed to elicit emotional responses. Video clips are particularly effective at inducing emotional responses in children (Schaefer, Nils, Sanchez, & Philippot, 2010). Children were presented video stimuli while wearing electrodes to examine their autonomic nervous system activity (ANS) through an electrocardiogram (ECG) – a measure of the electrical activity of the heartbeat. Parents and children viewed a 12-minute emotion induction video together, modified from previous research (Fortunato, Gatzke-Kopp, & Ram, 2013). Stimuli was taken from the *Lion King*, as it has been shown to induce emotional states in previous work (Fortunato et al., 2013; Von Leupoldt et al., 2007). The *Lion King* is advantageous compared to other video stimuli as it uses an animal character as the target stimuli, therefore eliminating the need for racial matching between stimuli and participants (Roberts & Levenson, 2006). The video included four emotional clips in chronological order from the movie; scary (Simba being chased by hyenas), sad (Simba's father dying), happy (characters singing

and dancing), and anger (Scar fights Simba). The task was modified from Fortunato et al. (2013) so that each emotional clip was of equal length (2 minutes). Before the emotional clips were presented, there was a 1-minute baseline that presented a neutral scene of swimming fish. Between each emotional clip a similar neutral baseline video was shown for 45 seconds to reduce emotional carry-over effects.

ANS data collection. During the emotional video task, ANS data was collected with Mindware Biolab 3.2 software (MindWare technologies, Ltd Gahanna, OH). Mindware mobile monitors were placed in a backpack for the child to wear while electrocardiogram (ECG) recordings (ECG) and respiration data were recorded continuously from the child throughout the visit. ECG recordings were attained with adjusted Lead II configuration involving three spot electrodes; one placed at the bottom-right rib, one on the top left clavicle, and a ground electrode located at the bottom left rib. Respiration rate data was recorded with four spot electrodes placements; two at the front of the body located at the top jugular notch, and below the sternum serving as receiving electrodes. Two electrodes serving as sending nodes were placed on the back.

ANS Data Processing. Heart rate and RSA were obtained through ECG and processed using Mindware HRV 3.2 software (MindWare technologies, Ltd Gahanna, OH). Inter-beat intervals were detected via peak-identification algorithms. HR reflects the mean time between successive R peaks in each 60-second epoch. From ECG, RSA was obtained using spectral analysis of the inter-beat intervals (IBI) recorded, using fast Fourier transformation on the IBIs, which were detrended, centered, and tapered with a Hamming window, to decompose heart rate time series into component frequencies. High-frequency heart rate variability was extracted to quantify RSA within frequency

bandwidths associated with respiration (.24-1.04 for children; Fracasso, Porges, Lamb, & Rosenberg, 1994). Values were natural log transformed. Data were analyzed using 60-second epochs, with no more than 15 seconds artifact for any one segment. All recorded epochs were visually reviewed and corrected for artifacts, missing or mismarked peaks. RSA baseline was the initial 60 second neutral clip. Each emotional clip had two 60 second epochs which were averaged together.

RSA reactivity was calculated as change scores by subtracting the initial RSA baseline from the RSA of each emotional condition. Positive RSA reactivity or change indicates RSA increasing from baseline (RSA augmentation). Negative RSA reactivity or change indicates RSA decreasing from baseline (RSA withdrawal). HR reactivity was calculated with the same method; mean HR during baseline was subtracted from mean HR during each emotional video.

Data analysis: All results were run with bivariate Pearson correlations, with an alpha level set at of .05. The Shapiro-Wilk test of normality indicated that distributions of all variables except the FSS ($p = .009$) and CTS ($p < .001$) were normal ($p > .05$). No extreme outliers (3x Interquartile range) were observed. Normality of residuals was inspected and met assumptions of normality.

First, we ran correlations between early life stress factors and RSA (baseline and reactivity) and heart rate (baseline and reactivity). Next, we examined correlations between RSA (baseline and reactivity) and HR (baseline and reactivity) with internalizing symptoms (depression and anxiety). Results were then run controlling for RSA baseline using partial correlations. Child sex and age were tested as covariates using partial correlations.

Missing Data. At the time of analyses for the current study, complete ANS data were processed for 19 children. Five children were missing ANS data due to an incident in which lab equipment was stolen. Twenty-six children had complete data for demographics and internalizing symptoms. The FRS and FSS was completed for 19 children due to parents not completing all visits of the study at the time of these analyses. There was no significant difference of demographics or family risk for children with missing data.

RESULTS

Preliminary Analyses

Child sex and age were tested as covariates and did not change the results so were not included here.

Question 1: *Does early childhood environmental risk, (low family income, lack of family support, reduced parental resources, and increased family violence exposure correlate with altered parasympathetic (RSA) and sympathetic (HR) nervous system activity during an emotion induction task?*

RSA associations

Baseline RSA. There were no significant associations between parent reports of SES (family income, family resources), family support, and domestic violence with children's baseline RSA levels for any of the emotion conditions (all p values > .05).

RSA Reactivity: Parent reported levels of family SES (income, family resources) family support, and domestic violence were not significantly associated with RSA change

for happy, angry, sad, or scary videos (all p values $> .05$). See Table 2 for results. As a next step in analyses, baseline RSA was included as a covariate in the model. There were no significant associations between RSA reactivity and risk factors with baseline RSA in the model.

HR associations

Baseline HR. There were no significant associations between parent report of SES (income, family resources), family support, or domestic violence and baseline HR levels (all p values $> .05$).

HR Reactivity. There were no associations between family income or family support on children's HR reactivity during any of the emotion conditions (all p values $> .05$). Parent report of family resources was significantly, positively associated with child HR reactivity during the sad video ($r = .74, p = .038$) and, at a trend level, during the happy ($r = .63, p = .095$) and angry ($r = .65, p = .083$) videos. As demonstrated in Figure 1, as family resources increased, HR increased in response to the sad video. Parent report of domestic violence was significantly, negatively associated with child HR reactivity during the scary video ($r = -.67, p = .05$), and a trend for significance in the happy ($r = -.59, p = .09$) video. As shown in Figure 2, as domestic violence increased, HR slowed down in response to the scary video. See Table 2 for results.

Question 2: *To what extent is variability in parasympathetic (RSA) and sympathetic (HR) nervous system activity during an emotional induction task significantly associated with children's risk for internalizing symptoms?*

RSA associations

Children's RSA baseline and reactivity to each emotion condition were then examined in association with parent reports of child depression and anxiety symptoms.

Baseline RSA. RSA baseline was not associated with depression or anxiety symptoms (all p values $> .05$).

RSA Reactivity. RSA reactivity during the sad video was significantly, positively associated with child depression symptoms ($r = .619, p = .014$) as shown in Table 2. When baseline RSA was entered as a covariate in the model, associations between RSA reactivity in the sad condition ($r = .62, p = .014$), angry condition ($r = .74, p = .003$), and happy condition ($r = .63, p = .016$) emerged as significant, See Table 3. As depression symptoms increased, RSA reactivity became more positive (indicating less RSA withdrawal); see Figure 3.

Anxiety symptoms were marginally associated with RSA reactivity to the sad video ($r = .51, p = .055$). When baseline was entered as covariate, RSA reactivity to the sad video was no longer significant ($r = .37, p = .191$), however RSA reactivity to the angry video became marginally significant ($r = .49, p = .07$).

HR associations

Baseline HR. Baseline heart rate was not significantly associated with parent reports of child depression symptoms but was associated with parent report of children's anxiety symptoms at a trend level ($r = .443, p = .098$).

HR reactivity. Heart rate reactivity to the sad ($r = -.63, p = .013$), happy ($r = -.67, p = .007$), and angry ($r = -.70, p = .004$) videos were associated with depression symptoms.

As depression scores increased, heart rate reactivity decreased; see Figure 4. Heart rate reactivity was not associated with children's anxiety symptoms (all $p > .05$).

DISCUSSION

This study examined the associations between early life stress, respiratory sinus arrhythmia (RSA), heart rate (HR), and internalizing problems in a low-income and ethnically diverse sample. Consistent with hypotheses, we found associations between specific risk factors, the availability of family resources and reported domestic violence, with alterations in children's sympathetic nervous system (HR reactivity) to sad and scary emotional stimuli. This study has also found associations between internalizing symptoms such as depression with both sympathetic (HR reactivity) and parasympathetic (RSA reactivity) responses to emotional stimuli. Results indicated that depression symptoms were negatively associated with heart rate reactivity and positively associated with RSA reactivity to sad, happy, and angry videos. Overall, findings point to links between early environment and reactivity to negative emotions, and these patterns may confer risk for depression symptoms.

Associations between early life stress and ANS activity

First, consistent with hypotheses, children's exposure to early life risk such as reports of family resources and domestic violence were associated with variability in children's sympathetic nervous system (HR reactivity) but not parasympathetic (RSA reactivity). Specifically, parent reported lack of resources was associated with reduced heart rate reactivity when children watched sad video clips. A lack of resources (broadly defined in this measure as a lack stability, abilities to support basic needs, time to support

basic child development) may interfere with parents' opportunities to help children develop effective emotional growth, regulation and competence. Children reared in homes that lack basic family resources may also be exposed to increased stress in the family environment, which may also compromise the development of effective emotional regulation strategies (De Wied, Boxtel, Posthumus, Goudena, & Matthys, 2009), and underlying physiological processes that support them.

Our findings also revealed that children reared by parents who reported higher levels of domestic violence, showed a significant decrease in heart rate reactivity when watching scary and happy video clips. Decreases or decelerations in heart rate is widely shown to correlate with increased attentional load (Porges & Raskin, 1969). The particular salience of the scary emotion within children that might be exposed to domestic violence at home may be indicative of the child's higher awareness in identifying threat in the environment. The combination of our findings may indicate that children who are reared in contexts that lack resources or include increased domestic violence exposure may be more attuned to emotionally evocative contexts.

Associations between Depression and ANS activity

Next, our study found a significant association between reductions in HR when children viewed sad emotional video clips, and increased depression symptoms. In the context of depression, depressed individuals are more likely to stay engaged to sad stimuli and disengage faster from happy stimuli (Levens & Gotlib, 2010). Sustained attention to the sad film would also fit with common symptoms of depression such as rumination and brooding over negative content.

Child depression symptoms were also associated with less RSA withdrawal in response to the sad video which may indicate some adaption or dysregulation of the parasympathetic response system. Previous work is conflicting on whether greater RSA withdrawal is related to higher (Boyce et al., 2001; Fortunato et al., 2013; Hastings et al., 2008b; Hinnant & El-Sheikh, 2009; Thayer & Lane, 2000) or lower (Gentzler et al., 2009; Schmitz, Kramer, Tuschen-Caffier, Heinrichs, & Blechert, 2011) internalizing symptoms. The greater RSA withdrawal or over-reactivity observed may contribute to more over-arousal, hypervigilance and anxiety or worry symptoms (Thayer & Lane, 2000) and lower RSA withdrawal may be more indicative restrictive capacity to adapt and respond to stress (Schmitz et al., 2011). Thus, our findings of lower RSA withdrawal in relation to depression rather than anxiety symptoms would fit in this understanding. Additionally, studies have used different tasks (cognitive challenges versus emotional tasks) that could influence the expected direction of the reactivity. Importantly, our findings of less RSA withdrawal to the sad video specifically are consistent with existing literature that less RSA withdrawal to sad emotional stimuli predicts greater risk for increased depression symptoms across time in youth and adolescents (Gentzler et al., 2009; Rottenberg, Gross, & Gotlib, 2005; Yaroslavsky et al., 2013). This is meaningful because the sad video was a particularly salient predictor compared to the other emotional stimuli, and sadness is a key symptom of depression. This connection highlights clinical significance in that children's autonomic response to sad stimuli could be helpful in predicting children at risk for depression and extends prior work to low income children.

Another explanation for this study's findings compared to previous literature on internalizing symptoms and RSA functioning may be due to differences in demographics and environmental contexts. According to a recent meta-analysis (Graziano & Derefinko, 2013) most studies were conducted with Caucasian and low environmental risk samples, whereas the context of this study's sample consisted primarily of African American and Hispanic children from high risk environments. A study conducted by Gray et al. (2017) had similar sample demographics (high risk, low SES, primarily African American) to the current study and received similar outcomes of increased parasympathetic activity in girls with internalizing symptoms, which may indicate the importance of context when evaluating physiological data. Environmental context plays an important role in understanding adaptiveness of physiological reactivity. For example, theoretical models such as Biological Sensitivity to Context (Boyce & Ellis, 2011) and Adaptive Calibration Model (Del Giudice et al., 2011) suggest that adaptations to physiology may be adaptive to survive and thrive in one's specific environment.

Limitations

As previously mentioned, the current report includes a small sample size, so results are preliminary and should be interpreted with caution. Furthermore, due to limited sample size, statistical power is decreased, so we were likely underpowered to detect findings with lower effect sizes. Parents were present during the emotional video task, which may have been a confounding factor as there is variability in how parents responded or interacted with their child during this task. Parent's own emotional response to the video or interaction with the child could have influenced the child's physiology (Feldman, Magori-Cohen, Galili, Singer, & Louzoun, 2011). All information on

socioeconomic status, environmental risk, and domestic violence were gathered through parent reports from high risk areas. Although parents were informed of research confidentiality with specific limitations, parents may have been hesitant to report sensitive information such as incidents of domestic violence due to skepticism of research institutions. Additionally, internalizing symptom information were gathered solely through parent reports. Gaining information from additional informants (teachers, other caregivers) may be useful for providing supplementary perspective in identifying child internalizing symptoms within more social contexts (child interacting with other children). However, for young children in preschool, many may not be in school, or have the capacity to self-report, so parent-report is considered a well-validated measure of internalizing symptoms. The present study was also cross-sectional and future work should utilize longitudinal methods to better understand the long-term implication and developmental trajectories of internalizing symptoms.

Despite limitations, this study has multiple strengths such as studying high-risk populations, which are particularly sensitive and difficult groups to recruit for research. Additionally, Hispanic children are underrepresented within studies, and expanding on physiological data and internalizing symptomology within this minority group may prove to be valuable to the literature and clinical settings. This study's methodology also takes apart multiple kinds of factors that are involved with high-risk environments to get a better understanding of what puts individuals the most at risk for internalizing symptoms. This may provide suggestions on what stressors to focus on in prevention or intervention efforts instituted to buffer the effects of high-risk environments on children's emotional

regulatory outcomes. This study may also continue to confirm that there are potential biological markers involved in depression in youth.

Future Research

For this project specifically, further collection of missing data and an increase in sample size will help clarify and confirm our hypothesis and current findings. Future work should also consider examining patterns of parent-child interaction and synchrony to understand how mothers and children's influence and co-regulate during emotional tasks. Including multiple informants for reporting domestic violence such as reports from the other partner or examining police reports may provide additional validity. Investigating other components of the ANS, such as pre-ejection period (PEP), that are more indicative of sympathetic activity may increase understanding of how these stress response systems function. The interaction between PEP and RSA will provide more detail to the literature on activation patterns within children from high risk environments.

In summary this research is important in demonstrating how variability in early life stress exposures shape underlying stress physiology and increase risk for internalizing symptoms. This sample involved families living in impoverished, underrepresented racial and ethnic minority groups, who varied in early life stressors. This is an important strength of the study, given that most research on ANS variability has been conducted in middle class, low risk children and families. In terms of implications, our findings point to key early markers on risk for internalizing symptoms which could improve earlier identification of internalizing symptomology and better screening practices for children from diverse groups.

Table 1.

Demographics

Variable	n (%)
	N = 31
Age (years)	
4	12 (39%)
5	10 (32%)
6	6 (19%)
7	3 (10%)
Child Race	
White	9 (29%)
Black	18 (58%)
Other	4 (13%)
Child Ethnicity	
Hispanic	12 (39%)
Non-Hispanic	19 (61%)
Child Sex	
Male	13 (42%)
Female	18 (58%)
Maternal Education	
Masters	2 (6%)
Some college	16 (52%)
Community college	4 (13%)
High School/ GED	4 (13%)
Some HS	4 (13%)
Jr high	1 (3%)
Income-Needs-Ratio, M(SD)	1.01 (.57)

Notes. Income-to- Needs ratio: 1.00 = 100% of federal poverty level

	Child's Age	Income	Child Sex	FSS	FRS	DV	Anxiety	Depression	RSA baseline	HR baseline	RSA scar y	RSA sad	RSA happy	RSA angry	HR scary	HR sad	HR happy
Income	.07																
Child Sex	-.13	-.18															
FSS	-.27	.06	-.28														
FRS	-.08	.29	.17	.69**													
DV	-.50*	.05	-.07	-.08	-.25												
Anxiety	.22	-.04	.12	.48*	.37	-.62*											
Depression	-.01	-.41	.17	.09	.09	-.10	.41*										
RSA _{baseline}	-.10	-.26	.25	-.56	-.44	.25	-.47	-.14									
HR _{baseline}	.01	.20	-.15	.21	-.06	-.16	.44	.31	-.66**								
RSA _{scary}	-.33	.19	.21	.25	.35	-.44	.39	.11	-.13	.13							
RSA _{sad}	-.23	-.36	.25	.07	-.18	.24	.51	.62*	-.43	.36	.52*						
RSA _{happy}	-.23	-.35	.28	-.58	-.60	.36	-.14	.28	.80**	-.49*	.23	.03					
RSA _{angry}	-.24	-.42	.24	-.61	-.47	.14	-.17	.26	.85**	-.54*	.11	-.08	.94**				
HR _{scary}	.36	-.08	.10	.07	.43	-.67*	.08	-.35	.16	-.28	-.32	-.55*	.00	.12			
HR _{sad}	.42	.45	-.24	.30	.74*	-.34	-.24	-.63*	.27	-.50*	-.33	-.85**	-.07	-.02	.60**		
HR _{happy}	.36	.35	-.15	.14	.63	-.59+	-.23	-.67**	.23	-.49*	-.29	-.73**	-.20	-.05	.54*	.85**	
HR _{angry}	.42	.25	.07	.24	.65	-.53	-.16	-.70**	.18	-.34	-.13	-.60**	-.22	-.23	.40	.72**	.74**

Table 2.

Correlations between study variables

* $p < .05$ (2-tailed); ** $p < .01$ (2-tailed)

Notes. HR= heart rate, RSA = respiratory sinus arrhythmia, FSS = Family Support Scale, FRS = Family Resource Scale, DV = domestic violence

Table 3.

Association between RSA reactivity and internalizing symptoms

Variables	RSA sad	RSA scary	RSA happy	RSA angry
Anxiety	.37	.30	.39	.49+
Depression	.63*	.07	.63*	.74*

Notes. This table includes baseline RSA as a covariate.

RSA = respiratory sinus arrhythmia

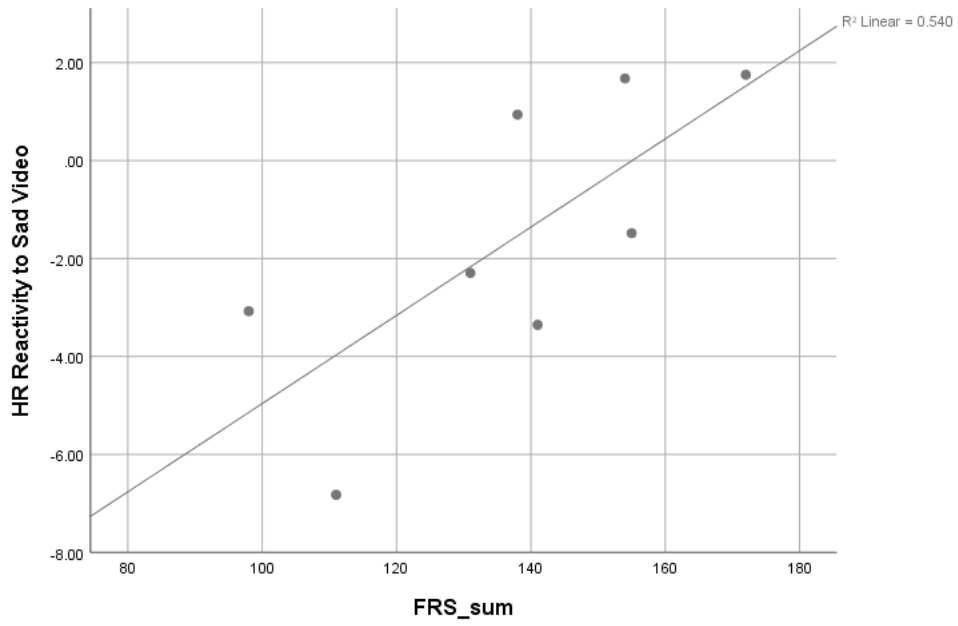


Figure 1. Correlation between Family Resource Scale and heart rate reactivity to sad video.

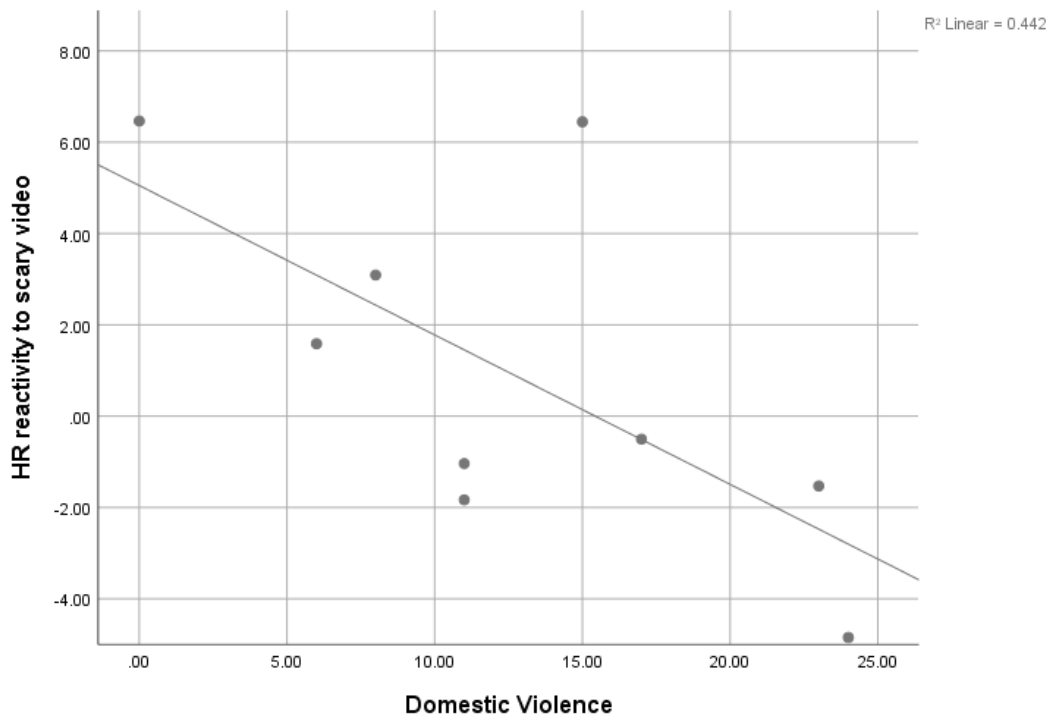


Figure 2. Correlation between domestic violence and heart rate reactivity to scary video.

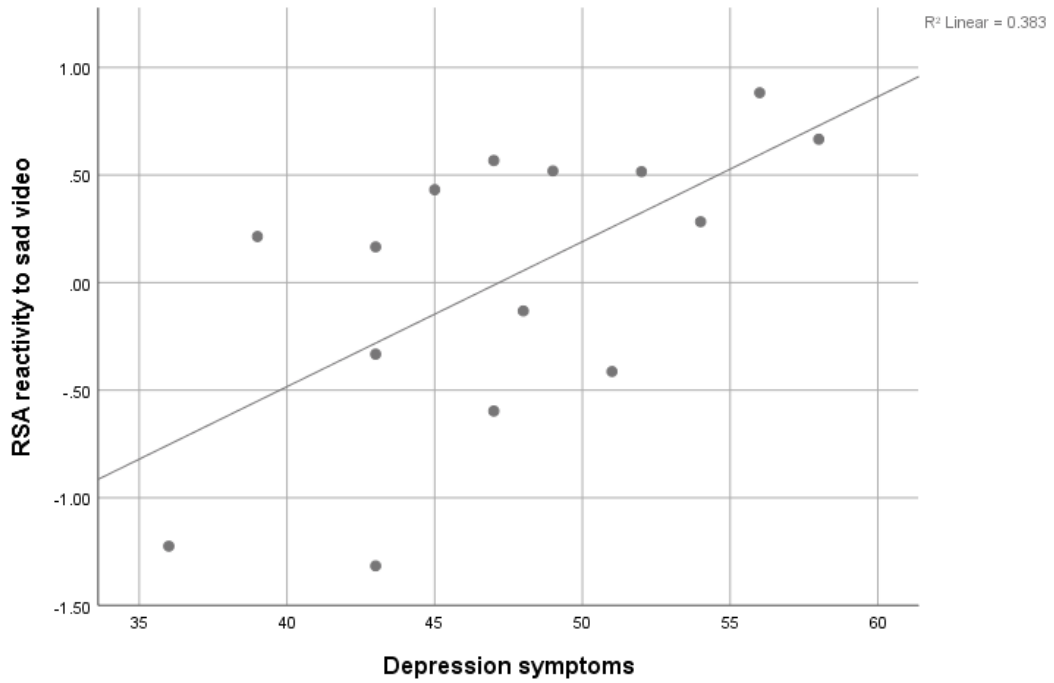


Figure 3. Correlation between depression symptoms and RSA reactivity to sad video.

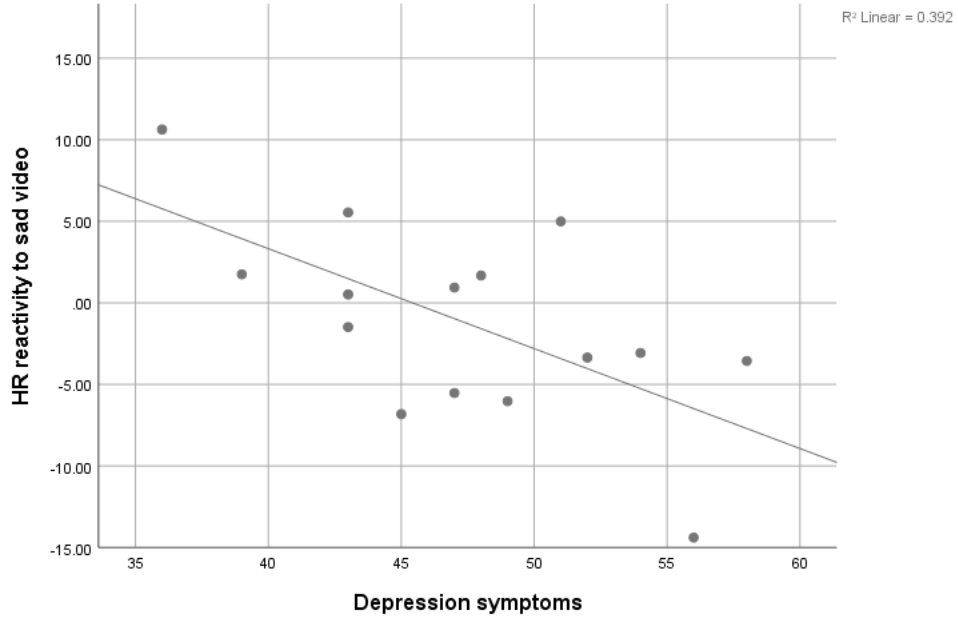


Figure 4. Correlation between depression symptoms and heart rate reactivity to sad video.

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