UNIVERSITY OF OKLAHOMA

GRADUATE COLLEGE

Emotional Attention Bias and Pupillometry Biomarkers as Mediators of Caregiving Effectiveness on Child Behavior

A THESIS

SUBMITTED TO THE GRADUATE FACULTY

In partial fulfillment of the requirements for the degree of

MASTER OF SCIENCE

By

JORDAN NORRIS

Norman, Oklahoma

2022

Emotional Attention Bias and Pupillometry Biomarkers as Mediators of Caregiving Effectiveness on Child Behavior

A THESIS APPROVED FOR THE

DEPARTMENT OF PSYCHOLOGY

BY THE COMMITTEE CONSISTING OF

Dr. Lauren Ethridge, Chair

Dr. Lara Mayeux

Dr. Michael Wenger

Acknowledgements

The current work is the culmination of so many bright, kind, and generous people. If not for the support and guidance I received, it would not have been possible. I like to think I am quite tenacious in my work, but I sit on the shoulders of giants. First and foremost, I am so grateful to Dr. Lauren Ethridge. Lauren has guided me in many ways over the past five years, but she gave me a home when I needed it most and words will never really convey the depth of my gratitude. I have an amazing space where I am challenged and free to challenge myself without fear. I am safe to fail and because of that I am safe to succeed. Thank you for your time, for all the feedback, for supporting my ideas, editing my writing, and for the financial contribution that allowed me to work primarily on this project.

I would like to show my gratitude to my committee for their support and recommendations. Both myself, and my work are better for it. I am appreciative of the support from both Dr. Lara Mayeux and Dr. Michael Wenger. I look forward to many more years in your respective orbits. A huge thank you to both Dr. Elizabeth Bard and Vicki Cook. I would not have gotten this far without your support and aid. Thank you for your kindness and for allowing me to take up space in your clinic.

To my lab, I thank you for all your help and support. Especially Emma Auger, who has helped and supported me in so many ways, whether that was coding or just being a friend who is as passionate about brains as I am. Thank you, your help and friendship mean so much to me.

Getting through any major graduate milestone requires much more than academic support. Special thanks go to all my friends and my cohort. I started my graduate education in the middle of a global pandemic, which was a wild time. My lovely Covid-Cohort was an

excellent support system full of beautiful, brilliant people that I am lucky to all call my friends. I am grateful for our laughs, and I am grateful to know that you are all there for me. I am so fortunate to be surrounded by such wonderful, passionate, and hardworking people. Thank you to Catherine, I appreciate you, your friendship, and all our adventures. I appreciate your unwavering personal and professional support. Thank you for listening to me when I talk about brains, being my science sound board, and putting up with all my opinions. I am better for knowing you. Thank you to those of you who put up with me talking about books, you know who you are. Thank you to Maddison North and Catherine, again, for helping with my presentation. Lastly, thank you to Elyse and Melissa for simply existing. I love you both dearly.

Finally, thank you to my family. There are not enough pages to tell you all the ways I love you and how grateful I am that you love me. You never fail to support my dreams and constantly reminded me that I am who I choose to be. Thank you for supporting me, always. This thesis stands as a testament to it.

Table of Contents

Acknowledgements iv

Abstract ix

Chapter One 1

Abstract 2
Background 4
Neural development 5
Prenatal Substance Exposure 6
Attention Patterns and Biases 9
Purpose and Research Objectives 13
Methods 14
Participants 14
Materials 15
Eye-Tracking 17
Procedure 17
Eye-tracking Tasks 18
Caregiver Task 18
Child Task 19
Areas of Interest (AOI) 20
Data analysis 20
Statistical Analysis 24
Results 26
Clinical Correlations 36
Child Exploratory Outcomes 40
Caregiver Exploratory Outcomes 41
Discussion 43
Primary Mediation Results 44
Anger Specific Attention Biases 47
Primary Correlations and Clinical Outcomes 48

Child Exploratory Outcomes 50
Caregiver Exploratory Outcomes 51
Conclusions and Future Directions 52
Limitations and Assumptions 53
Chapter Two 56

Abstract 57
Background 58
Pupillometry 59
Purpose and Research Objectives 60
Methods 61
Participants 61
Materials 62
Procedure 62
Pupillometry 62
Data analysis 63
Statistical Analysis 64
Results 65
Pupillometry Clinical Correlations 79
Child-Specific Pupil Results 80
Caregiver-Specific Pupil Results 80
Discussion 81
LC-NE Function Mediation Models 81
Conclusions 85
Limitations and Assumptions 85
References 87

Appendix A: Stimulus Specifications 99

Appendix B: Plots Visualizing the Conditional Effect of the Focal Predictor for Primary Mediation Models 101

Appendix C: Plots Visualizing the Conditional Effect of the Focal Predictor for Anger-Specific Attention Bias Mediation Models 104

Appendix D: Plots Visualizing the Conditional Effect of the Focal Predictor for Pupillometry Mediation Models 107

Appendix E: Plots Visualizing the Conditional Effect of the Focal Predictor with Emotion-Specific Caregiver Pupillometry Mediating 110

Appendix F: Plots Visualizing the Conditional Effect of the Focal Predictor with Caregiver Composite Mediating 113

Abstract

Early life adversity experienced during childhood and adolescence negatively impacts development with increased risk of poor health outcomes and early mortality, making child maltreatment a significant public health concern. Adversity exposure broadly impacts developmental trajectories and alters physiological processes supporting emotional processing and regulation (e.g., changes or dysregulation of hypothalamic-pituitary-adrenal (HPA) axis or autonomic nervous system function). Changes to neural physiology resulting from adversity exposure dynamically influence the way children engage with the environment and the way the environment continues to influence physiology and behavior throughout development. Largely, trauma-centered research has focused primarily on children and child outcomes without factoring in caregivers as a developmental context. Caregivers are important for the development of emotion regulation and emotion processing skills with decreased emotional literacy negatively impacting the relationship between early life stress and typical behavior. An eye-tracking study employing an emotion identification task was conducted on caregiver-child dyads to assess two potential biomarkers. Attention biases exhibited by children with prenatal substance exposure in response to emotionally valenced social stimuli were evaluated as a potential biomarker for emotional processing dysregulation. Results suggest physiologically characterized emotional literacy in the caregiver mediates the relationship between negative emotion processing and significant behavior problems in the child. Problematic behaviors are likely rooted in physiological shifts and changes to autonomic nervous system function may play a role in the relationship between adversity exposure and child behavior. Child pupillometry served as a biomarker of changes to locus coeruleus and norepinephrine reactivity in response to negatively valenced emotional stimuli. Results demonstrated caregiver emotional literacy partially mediated

the relationship between child pupillometry and externalizing behaviors. Broadly, results demonstrate the important role of caregivers as mediators of child emotional processing and emotion regulation for children with significant life adversity.

Chapter One

Emotional Attention Bias as a biomarker of Emotional Processing and Literacy on Child Behavior

Keywords:

prenatal substance exposure; child attention biases; child behavior problems; trauma

Abstract

Adverse childhood experiences (ACEs) are broad categories of trauma experienced by children and adolescents prior to the onset of young adulthood. Adversity exposure and maltreatment early in life are related to later negative life outcomes via the interplay between trauma factors and developmental processes. ACEs are thought to disrupt emotional processing and regulation skills with limited work addressing the physiological mechanisms by which these disruptions occur. Prenatal factors, like prenatal substance exposure (PSE), reflect a potential source of physiological abuse with implications for emotional processing, executive functioning, and behavioral inhibition capacity (e.g., impulse control). Predicting consequences of ACEs results in a focus on innate child outcomes with the functional abilities of the caregiver and their behavioral impact on the child potentially overlooked. Specifically, children are the focus of trauma in developmental research with limited focus on how caregiver emotional literacy and emotion related behaviors affect child developmental outcomes. Eye-tracking provided a non-invasive measure of physiological function for testing the mediating effect of caregiver emotional processing to address caregiver functional ability and emotional-related behaviors in relation to child outcomes at a physiological level, reflecting a novel approach. Thirty caregiver-child dyads participated in emotion identification tasks using eye-tracking technology to record gaze-point and search strategy data. All children had confirmed or suspected prenatal alcohol or drug exposure. Adult participants (N = 30; Mean age = 42.34 years, SD = 7.68; 25 females) were instructed to search for a specific target emotion from six faces expressing emotions ranging from positive to negative (i.e., happy, neutral, sad, anger, fear, and disgust). The child task differed with child participants (N = 30; Mean age = 5.10 years, SD = .92; 4 Females) only required to identify the face that was different, where 5 faces expressed a neutral emotion and

one expressed either sad, fear, or anger. Eye-tracking measured attention allocation to negatively valanced stimuli in children to establish physiological correlates of attention biases with trauma exposure, externalizing behaviors, and caregiver emotional processing capacity. Caregiver emotional processing mediated the relationship between child attention bias and externalizing behaviors demonstrating that caregivers play a role in the behavioral environment of the child. Using attention bias as a biomarker of emotion processing will enrich current literature on trauma and the mechanism by which it disrupts physiological processes, and provide insight into physiological mechanisms underlying current therapeutic inventions by providing a means to test efficacy and push for more individualized interventions based on physiological outcomes.

Background

More than 60% of American adults are exposed to at least one adverse childhood experience (ACE) during their youth. ACEs are defined as abuse, neglect, or traumatic experiences occurring prior to the age of 18, and are characterized broadly to encompass most external experiences resulting in child trauma (Merrick et al., 2018). Formally, ACEs fall into one of three categories. Abuse and neglect represent two types of adversity exposure and can either be physical or emotional, with physical abuse including any form of physical violence or sexual abuse toward the child. Other traumatic experiences fall into the category of household dysfunction, broadly defined as disruptions in a child's environment that result in traumatic experiences. Household dysfunction is further subdivided into 1.) divorce or voluntary removal of an adult/caregiver from the household environment and nonviolent conflict, 2.) domestic abuse or violent conflict in the home between adults/caregivers or an adult/caregiver and a sibling, 3.) mental illness in adults/caregivers or siblings, 4.) substance abuse where others in the child's environment are abusing drugs, alcohol, or both, and 5.) incarcerated relatives or forced removal of adults from the child's environment (Felitti et al., 1998; Font & Maguire-Jack, 2016).

Current evidence supports that the impact of ACEs on adult health status is strong, widespread, and cumulative. Early life adversity exposure has a cascading effect that inevitably increases the risk for developmental psychopathology and early mortality. Specifically, early life adversity (i.e., ACEs) is associated with 5 out of the top 10 leading causes of death in the United States including cardiovascular disease, cancer, respiratory diseases, diabetes, and suicide (Felitti et al., 1998). Adversity exposure forms a foundation comprised of neurodevelopment disruptions leading to later social, emotional, and cognitive impairment. Disruptions to psychosocial

mechanisms, emotional processing, or impaired cognitive functions increase the likelihood of individuals adopting health risk behaviors and later experiencing social problems, disability, and/or disease. Mechanisms underlying biological/neural embedding of trauma and early life adversity remain poorly understood despite known associations between adversity experience and negative life outcomes. Potential mechanisms include trauma acting upon neural cytoarchitecture via stress pathways (e.g., HPA axis and locus coeruleus-norepinephrine system reactivity) resulting in changes in volume and functional shifts in limbic structures (Anda et al., 2006; Herzog et al., 2018; Thomas et al., 2016). Stress effects are assumed to mediate the relationship between ACEs and development of disorders and neural alterations. However, the full effect of adversity vulnerability on negative life outcomes requires an understanding of ACE type and timing of adversity exposure before fully drawing meaningful conclusions (Herzog et al., 2018).

Neural development

Arguments for biologically defined adversity sensitivity and increased risk related to ACE timelines stem from work supporting developmental sensitive periods as times during neural development when children are thought to be more susceptible to environmental factors (Bick and Nelson, 2016; Reh et al., 2020). While brain plasticity is dynamically regulated across the human lifespan, sensitive periods are thought to span from early infancy to early adolescence representing a window of rapid and vulnerable neural plasticity responses (Reh et al., 2020). Evidence examining the neuroplasticity of trauma circuits indicates that the sensitive window is more extensive than prior thought and reaches into late adolescence, peaking at the age of 10 (Stevens et al., 2018). Circuitry for emotional processing and other higher executive functions are complex with sensitive periods occurring later than sensitive periods for more basic neural

circuits (Bick and Nelson, 2016; Fox et al., 2010). Trauma experienced during the large sensitive window can shift development toward neural outcomes associated with risk placing children with adversity exposure on a path to negative life outcomes.

Sensitive periods tell us about the role of plasticity and vulnerability during development with recent evidence suggesting emotional processing capacity plays a role in altering the interaction and risk for negative outcomes associated with experiencing ACEs. For example, attention biases away from negative emotions (e.g., anger and fear) leading to greater levels of emotional insecurity following exposure to interparental conflict (Davies et al., 2020a). Emotional insecurity is driven by emotional regulation and underlying emotional processing abilities where the child can control stress arousal and emotional responses. Self-regulation is an important aspect of emotion processing and regulation impacted by environmental situations, social relationships, and personal emotion regulation drives of the child (Thompson & Calkins, 1996). Recent work suggests that prenatal adversity exposure in the form of fetal exposure to substances *in utero* may result in reduced emotional regulation capacity via alterations to the brain (Morie et al., 2019; Eiden et al., 2016).

Prenatal Substance Exposure

Human development is complex in terms of external factors influencing physiological mechanisms underlying typical development. The prenatal environment is an important neurodevelopmental context in which negative prenatal factors result directly in negative infant developmental outcomes. Specifically, prenatal substance exposure (PSE) reflects embedded physiological trauma stemming from caregiver behavior acting as a significant barrier to healthy infant development and a predictor of later ACEs (Morie et al., 2019; Roos et al., 2021). Most substances (i.e., drugs) cross the placental barrier affecting fetal development to varying degrees

depending on the substance use timeline during the pregnancy and the frequency of use (Ross et al., 2015). Known pharmacokinetics show the tendency for drugs of abuse to target major protein and molecular functions resulting in deleterious developmental outcomes. Benefits to exploring substance exposure as a form of adversity exposure are the known pathways and mechanisms of physiological disruption though the adversity effects are often severe (Eiden et al., 2016; Morie et al., 2019; Ross et al., 2015).

Drugs typically abused during pregnancy include cocaine, amphetamines, alcohol, and opioids for which mechanisms of developmental disruption have been described. Cocaine impacts dopamine (DA), noradrenergic, and serotonin systems by blocking monoaminergic transporters. Prenatal cocaine exposure (PCE) potentially disrupts the development of DA-systems, as the DA-system develop early in vertebrates rendering fetuses sensitive to exogenous cocaine influence. PCE tends to result in alterations to neuroanatomy and disruptions to cognitive function with a dominant developmental phenotype of *in utero* cocaine exposure resembling symptoms of ADHD, though outcomes are typically quite varied (Eiden et al., 2016; Morie et al., 2019; Thompson et al., 2009).

Amphetamines are monoaminergic in nature similarly to cocaine but the effects of amphetamines (e.g., methamphetamine) *in utero* are distinct from cocaine. Amphetamines reverse the effects of monoamine transporters increasing neurotransmitter availability in the synaptic cleft. Further, amphetamines reduce the reuptake and degradation of neurotransmitters in the synaptic cleft amplifying the effects of monoamines on the post-synaptic neuron. Children exposed *in utero* to amphetamine and methamphetamine show increased stress, decreased arousal, and increased risk for long-term cognitive deficits (e.g., attention, memory, visual motor integration, etc.). Additionally, prenatal amphetamine exposure results in neuroanatomical

changes to striatum and hippocampal volume with reported decreases in D_2 receptors. Use of multiple substances during pregnancy complicates the clinical presentation of PSE (Morie et al., 2019; Thompson et al., 2009).

Alcohol is a known teratogen with prenatal exposure effects including disruption of numerous histogenic processes via disruption of intracellular processes (e.g., growth factor receptor signaling via degradation to plasma membrane integrity). Neural tissue is often damaged by exposure to alcohol *in utero* and proliferation disruptions resulting in reduced neuron formation and size. Prenatal alcohol exposure (PAE) results in alterations to major functional networks through alterations in microstructure (i.e., cytoarchitecture) (Roos et al., 2021). Neurodevelopmental outcomes associated with prenatal alcohol exposure are relatively severe, including intellectual disability and deficits to cognitive, motor, and basic neural functions (e.g., attention). Despite severe consequences, the clinical presentation tends to be less complex than PSE because alcohol is more frequently used as a solitary drug compared to other substances of abuse (Morie et al., 2019; Thompson et al., 2009).

Exposure to opioids *in utero* results in similar neural outcomes associated with PAE in terms of teratogenic effects with similar cellular and functional consequences. Societally, prenatal opioid exposure (POE) is of particular concern with an estimate of at least 1 in 5 women using an opioid during pregnancy across high-income countries resulting in negative infant neurocognitive and developmental outcomes including reduced motor abilities (Yeoh et al., 2019). Animal studies show POE decreases neurogenesis and neurotransmitter levels, increases cell death (i.e., apoptosis), and is typically accompanied by alterations in myelination. Neuroimaging studies found alterations to major neural structures and microstructure of the developing brain and functional imaging showed altered function networks. Namely, amygdala

connectivity is altered in POE. Functional connectivity differences of the amygdala noted in POE suggest differences in regulation of emotions, stress, and associated behaviors (Merhar et al., 2021; Radhakrishnan et al., 2020)

In summary, PSE has varying effects depending on the type and frequency of substance use but broadly results in negative cascading developmental effects predictive of adverse life outcomes. *In utero* exposure to substances causes persistent deleterious effects on cognitive function, particularly functional networks, attention, and regulatory control, in infancy and early childhood that continue to exert influence over neural and behavioral development into adulthood (Eiden et al., 2016; Morie et al., 2019; Nygaard et al., 2016). Elucidation of physiological mechanisms of adversity exposure, with specific attention to PSE generally, and the capacity to explore intervention efficacy are both necessary steps in addressing trauma and physiologically/biologically defining ACE exposure.

Attention Patterns and Biases

Attention is a cognitive function broadly affected by both ACEs and exposure to substances *in utero*. Infants exposed prenatally tend to exhibit decreased attention, altered attention patterns, and increased attention related problems (Davies et al., 2018; Morie et al., 2019; Nygaard et al., 2016). While PSE is an inherently highly detrimental form of adversity exposure, other types of adversity exposure may compound with developmental outcomes associated with the exposure to substances *in utero*. Risk factor accumulation may contribute to more extreme clinical profiles compared to independent presentations of PSE or adversity exposure (Koponen et al., 2020).

Social and relationship factors like caregiver-child attachment also contributes to attention patterns noted in children with ACEs (Vandevivere, et al., 2014). Infants are

biologically predisposed to form attachments with caregivers in their immediate environment and typically exhibit initial signs of attachment within the first year of life. The capacity of the caregiver to respond appropriately to the mental state of the child is an important social factor that promotes shared caregiver-child experiences. Shared experiences are important for positive developmental outcomes as they reflect internalizations of social experiences and shared emotional states that support the ensuing development of self-regulation in the child (Osher et al., 2020; Thompson & Calkins 1996). While mechanisms of attachment are not well defined, it is understood that internalization of social stimuli plays a role in attachment formation and style. Insecurely attached children exhibit attention biases from attachment information and socially relevant stimuli (Vandevivere et al., 2014).

Household dysfunction also contributes to shifts in attention patterns. While interparental conflict as a form of adversity does not guarantee insecure attachment in the child, patterns of attentional shifts away from a negative stimulus, particularly an angry face, are similar in insecurely attached children (i.e., avoidantly attached) and children from high conflict homes (Davies et al., 2018; Vandevivere et al., 2014). Additionally, adversity exposure is a seemingly strong predictor of insecure responses (Davies et al., 2020a). Overlap in attention patterns from PSE, ACEs, and attachment perspectives provides evidence in support of a fundamental neural source from which insecurity and deregulated emotional processing stems. Attention bias away from negative stimuli is likely a behavioral compensatory effort to cope with negative social information acting on stress or intrinsic vulnerability (Luecken et al., 2004). Alterations to emotional processing pathways result in insecure relationship formation and decreased emotion regulative capacity which subsequently informs future vulnerability to adversity (Davies et al., 2018; Davies et al., 2020a; Osher et al., 2020). Further, child removal is often a concern in cases

of confirmed and suspected PSE. Decisions to remove children from their caregivers/homes are not made lightly and often serve in the best interest of the child to promote future safety and well-being. Removal may mitigate known ACE risk, but removal during sensitive periods undermines the child's sense of security and attachment, and potentially results in long-term emotional and psychological consequences driving the need to develop coping mechanisms (e.g., attention biases away from negative affective stimuli) (Goldsmith et al., 2004).

Children with known ACEs tend to exhibit attentional shifts with increased rates of attention related conditions (Nygaard et al., 2016). Interparental conflict (i.e., nonviolent conflict) is an ACE under household dysfunction that leads to poor behavioral outcomes in children, such as internalizing and externalizing problems in the extrafamilial setting (Buehler et al., 1997; Davies et al., 2018). Previous work has shown that children from high conflict homes monitor facial expressions of emotion differently compared to children from low conflict homes (Briggs-Gowan et al., 2015; Davies et al., 2018; Schermerhorn et al., 2015**)**.

High conflict renders the home environment an at-risk setting for children resulting in emotional insecurity from pervasive exposure to negatively valenced social situations, regardless of whether the conflict is aimed at the child, leading to increases in negative reactivity (Davies et al., 2020b; Felitti et al., 1998). Emotional insecurity intensifies negative or pessimistic representations of conflict with implications for the child's sense of well-being (i.e., the child assumes that negative social stimuli in the external environment will have a negative impact on their well-being). Evidence suggests these pessimistic representations of household disruptions influence the transmission of interparental conflict to the child in the form of negative representations of the home environment (Davies et al., 2018). Recent work found the more aversive components of conflict exposure predicted emotional insecurity in children who attend

more strongly to negatively valenced emotional stimuli with increased attention related to internalization of social threat (Davies et al., 2020a). Transmission of negative affect may be unintentional and suggests diminished emotional literacy in caregivers that live in dysfunctional home environments.

Broadly, caregivers contribute to healthy emotional processing and the development of self-regulatory skills (Osher et al., 2020). Caregivers equally contribute to the development of maladapted emotional processing and regulatory skills (Erriu, 2017; Wolicki et al., 2021). It is therefore important to consider individual factors relating to previously experienced ACEs and other psychosocial factors in the caregiver as risk factors for trans-generational trauma. Early maternal trauma exposure (i.e., before the age of five) and psychopathological risk represent factors that impact mother-infant interactions and eventually pose a risk for trauma transfer and risk for later psychopathology (Erriu, 2017; Tambelli et al., 2015). Namely, caregiver trauma and stress affect relational quality and early infant health behaviors (e.g., feeding behaviors) (Ballarotto, 2016). Caregiver-child relationship interruption (i.e., failure to provide a stable and nurturing relational environment) contribute to risk for both chronic and toxic stress in the child accompanied by a reduction in stress and emotional regulation capacity (Bethell et al., 2017).

Current research on attention in maladapted caregiver-child relationships has focused on child outcomes, with little understood about how caregivers in at-risk settings allocate attention to the valence of socially relevant stimuli, such as facial expressions, although behavioral research supports reduced emotional literacy in caregivers in high conflict households or that have a history of abuse (Bozkurt et al., 2020; Merrick et al., 2018; Lieneman et al., 2017). Trauma does not exist in a vacuum making it necessary to understand the effects of childhood trauma exposure to prevent negative life outcomes in children. Caregivers are an important

developmental context elevating the need to understand the extent that trauma has potentially impacted or shaped their cognitive state, emotional processing strategies, and their ability to engage with their child in a healthy manner in situations of child adversity exposure (Wolicki et al., 2021).

Trauma has known effects on human physiology via activation of stress-related physiological systems. However, how trauma-related shifts in physiology ultimately contributes to behavioral outcomes is modified by bidirectional caregiver-child relationships and reflects an understudied area of development. A physiological measure of emotional processing may provide novel insight into the biological and cognitive mechanisms underlying negative neurodevelopmental outcomes and potentially lead to novel advancements in therapeutics designed specifically to address those biological constructs in cases of maladaptation or trauma. Outcomes from electroencephalography (EEG) suggest a physiologically rooted link between neural physiology, parenting, and child behavior. Bidirectional physiological responses theoretically could underlie the connection between negative acute caregiver-child interactions, attachment, and the presence of internalizing/externalizing behaviors (Atzaba-Poria et al., 2017).

Purpose and Research Objectives

Eye-tracking provides a novel opportunity to noninvasively address physiological aspects of attentional bias across caregiver and child dyads in a way that is more efficient and scalable than EEG. Eye-tracking helps elucidate the physiological underpinnings of biased attention in the presence of negatively valenced emotional stimuli within maladaptive caregiver-child relationships.

In the current study, we aim to use eye-tracking to measure attention allocation to negatively valenced stimuli in children to assess correlates of attention biases with trauma

exposure, and externalizing behaviors. We hypothesized that biased attention away from negatively valenced emotional stimuli would correlate directly with trauma exposure, externalizing behaviors, and cognitive/motor complications associated with PSE. We hypothesized further that decreased attention to anger would predict decreased security in the caregiver-child relationship and increased externalizing behaviors in the child. Finally, we hypothesized that caregiver facial emotional processing strategies as a composite would act as a mediator for the relationship between child emotional processing strategies and predict child externalizing behavior as a reflection of reduced emotional literacy in the caregiver.

Methods

Participants

Caregiver-child dyads (N = 30) with children between ages 3 and 7 (M = 5.10, SD = 0.920; Range: 3.10 - 6.91) were recruited from the Child Study Center at the University of Oklahoma Health Sciences Center (OUHSC), which is Oklahoma's only center for trauma intervention and research. Families were patients at A Better Chance Clinic (ABC) and children either had a confirmed or suspected history of prenatal substance exposure at the time they were recruited to participate. Eighty-one percent of the current sample had confirmed substance exposure and 38.1% had suspected exposure (some participants had confirmed exposure with suspected use of additional substances during the prenatal period) (**Table 1)**. Most children in the current sample were exposed to multiple substances during the prenatal period (N = 27) making it difficult to assess substance-specific outcomes. In addition to substance exposure, some children carried a diagnosis of FAS (N = 4) and presented with various neurodevelopmental disorders or developmental complications as a result of substance exposure. Adults (M = 42.34, SD = 7.68; Range: 27.29 – 59.14) were either the biological parent to the child, a biological relative acting as legal guardian, foster parent, or an adoptive caregiver (hereafter "caregivers").

All caregivers have maintained a relationship with the child for a minimum of 6 months (i.e., has known the child for longer than 6 months and contributes to care) to ensure the caregiver-child relationship was established with time allowed for caregiving behaviors to affect development.

Table 1. Substance List with Suspected and Confirmed Exposure Percentage and Sample Size

Substance	N	Percent Exposed
Alcohol		
Beer	2	6.7%
Liquor	4	13.3%
Unknown	5	16.7%
Substances		
Nicotine	12	40%
Marijuana (THC)	17	56.7%
Opiates	8	26.7%
Cocaine	3	10%
PCP	2	6.7%
Methamphetamine	20	66.7%
Amphetamine (other)	4	13.3%

Note. List of confirmed or suspected substances with the number and percentage of participants with reported exposure/suspected exposure.

Materials

At the time of intake, dyads completed a series of self-report or clinician rated clinical assessments including a trauma screener, Behavior Assessment System for Children- Third Edition (BASC-3), and Wechsler Preschool & Primary Scale of Intelligence – Fourth Edition (WPPSI-IV). Caregivers also completed the Eyberg Child Behavior Inventory (ECBI) when they reported externalizing child behaviors they judged to be problematic.

Eyberg Child Behavior Inventory (ECBI) is a behavior specific caregiver-rated report of externalizing behaviors that assesses current frequency (intensity) and severity (problem) of disruptive behaviors in the home and educational setting. Raw scores from the ECBI were used for analyses. Caregivers were asked to rate the frequency children exhibited item behaviors on a

scale from 1 ("*never*") to 7 ("*always*") and report whether those behaviors are a problem for the caregiver on a binary scale (i.e., "*is this a problem for you*" Yes/No). The ECBI is specifically provided if caregivers report problematic behaviors and aims to distinguish normal behavior problems from clinically significant behavior problems (i.e., conduct disorders) (Boggs et al., 1990). Only caregivers that reported externalizing symptoms to the clinical team were asked to report on ECBI. T-scored ECBI responses showed acceptable internal consistency ($\alpha = .71$).

Behavior Assessment System for Children – Third Edition (BASC-3) is a comprehensive behavior specific caregiver-rated report that aims to assess behavior and emotions broadly (Reynolds & Kamphaus, 2015). A 4-point response scale is provided for all items and caregivers were asked to rate whether their child never, sometimes, often, or almost always exhibited the item behavior (i.e., scored: 0 = *never*, 1 = *sometimes*, 2 = *often*, 3 = *almost always*). Composite scores were generated by summing across items belonging to composite scales (i.e., externalizing behaviors, internalizing behaviors, adaptive skills, and behavioral symptoms index) and standardizing for age. The BASC-3 also has several content scales that index more specific behaviors (i.e., anger control, bulling behavior, developmental social disorders, emotional self-control, executive functioning, negative emotionality, and resiliency). All caregivers were asked to provide BASC-3 responses.BASC-3 responses showed good internal consistency for subscale scores ($\alpha = .83$) (Wilder et al., 2003).

Wechsler Preschool & Primary Scale of Intelligence – Fourth Edition (WPPSI-IV) is a clinician rated cognitive assessment aiming to measure cognitive development in young children through preschool-ages (Wechsler, 2012). All children received age-appropriate WPPSI-IV testing which provided a full-scale IQ index (FSIQ) that included three subscales (i.e., verbal comprehension index (VCI), visual spatial index (VSI), and working memory index (WMI)).

Two additional subscales were calculated for children over the age of 4 to index fluid reasoning and processing speed (i.e., FRI and PSI, respectively).

Caregivers reported demographic information for the child and themselves directly to clinical teams. Caregivers also reported information about the biological parent (if available) and the current use of that information is limited to child trauma exposure during biological caregiver custody.

Eye-Tracking

A Tobii T120 eye-tracking system measured attention with participation occurring at the time of intake after clinical testing was completed. Stimuli were sourced from the NimStim Face Stimulus Set, a research-quality database of ethnically diverse male and female adult actors displaying seven emotional expressions each (Tottenham et al., 2009). Sex and race of each actor were balanced across stimulus types such that sex was equally represented 50/50 and race reflected the national percentage of the US as reported on the 2019 U.S. census (Bureau USC, 2019). Stimuli development and presentation was conducted via Tobii Pro Studio and run through the Tobii T120 eye-tracking system integrated into a 17-in TFT monitor. Infrared sensors on the eye tracker used pupil center corneal reflection to track the center and size of the pupil and corneal surface reflection to track the position of participant gaze from both eyes (Tobii Pro AB, 2012).

Procedure

Obtainment of formal written consent occurred at the time of clinic intake along with clinical assessments and demographics. Verbal consent was obtained prior to eye-tracking participation from the caregiver for both caregiver and child participation. Children provided verbal assent and were allowed to discontinue participation if uncomfortable. Eye-tracking

recordings took place in a dimly lit room approved for use by the Child Study Center, as the infrared sensors on the Tobii Eye-Tracking system work best in low-lighting (Tobii Pro AB, 2012*b*). Caregivers were asked if they or the child had any eye conditions that they were aware of (e.g., astigmatism, cataracts, glaucoma, etc.) and whether they wear corrective lenses for visual deficits or eye-conditions. All participants had either normal or corrected to normal vision.

Eye-tracking Tasks

Eye gaze location was first calibrated by asking the participant to look at the screen and confirming that the sensors on the eye-tracking system detect both eyes. Calibration continued if both eyes were detected with the participant following a red dot as it moves through five points across the center, edges, and corners of the screen.

Caregiver Task

Adult caregivers were asked to sit in a comfortable chair with their eyes 60 cm from the Tobii computer monitor measured using the Tobii calibration tools. Data collectors provided caregivers with a wired mouse connected to the stimulus delivery laptop that was not active until the task began. The task was explained to the caregiver, indicating that their job was to continually look at the screen without moving their head, and any time a white cross was on the screen (i.e., central fixation point) to orient their eyes to the center of the screen and look at the cross until the next trial begins. When presented with face stimuli (i.e., a trial) the caregiver was to find the target emotion indicated by the target word presented in the center of the screen and use the mouse provided to click on their choice.

The caregiver task contained 5 practice trials to acclimate participants to the task and 120 test trials with 20 trials for each target emotion, randomly interleaved. Average participation time took 13.5 minutes (M = 13.71, SD = .48) not including calibration. Each trial was 3500ms and

consist of 6 adult faces in a circle equidistant from a central fixation point all from one actor exhibiting standard emotional expressions. Trials were 880 pixels by 880 pixels with a resolution of 22 pixels per centimeter. Actor images were 3.32 cm wide with a height of 5.01 cm (see Appendix A, Figure A1). Each of the six actor images displayed a different emotion (happy, disgust, sad, angry, fearful, and neutral), with a word representing the target emotion for identification located in the center (i.e., replacing the central fixation point). Central fixation points were displayed between all trials for 2000ms to facilitate reorientation to the center of the screen.

Child Task

Child participants sat in a comfortable chair with their eyes 60 cm from the Tobii computer monitor measured using the Tobii calibration tools. Chair and table height were adjusted to ensure comfort and to allow participant's feet to contact the floor while maintaining the distance from eyes to screen (60cm). The task was explained to the child, indicating that their job was to continually look at the screen without moving their head (while holding their head as still as possible) and to orient their eyes to the center of the screen any time a smiley face emoji (i.e., central fixation point) was on the screen. When presented with face stimuli (i.e., a trial) their job was to find the face expressing the emotion that was different from other faces on the screen, but they were not required to make any response when they find the target face. The purpose of this direction is to orient the child toward faces displaying a negative emotion while allowing them to internally regulate the amount of time they spend attending to the negative emotion once it has been found.

The task for child participants contained 5 practice trials and 60 test trials (20 trials for each emotion, randomly interleaved), for an average participation time of 6.5 minutes (M = 6.20,

SD = .00) not including calibration. Trials were 880 pixels by 880 pixels with a resolution of 22 pixels per centimeter. Actor images were 3.32 cm wide with a height of 5.01 cm (see Appendix A, Figure A2). The intertrial period contained a smiley-face emoji for central fixation to improve participation. Each trial consisted of 6 adult faces in a circle equidistant from where the fixation point was, all from one actor exhibiting standard emotional expressions. The facial emotion identification task replicated prior methods, with 5 neutral and 1 negative face (sad, angry, fearful) for the participant to identify (Davies et al., 2018). Central fixation points were displayed between all trials for 2000ms to facilitate reorientation to the center of the screen.

Areas of Interest (AOI)

Predefined AOIs determined the speed of detection for target faces, visual search patterns, ratio of time spent viewing eyes, and subsequent length of attention to the face once detected using Tobii Studio software. AOIs were subdivided into eye and mouth areas to determine the primary face areas used by each participant group (caregivers and children) during visual search for the target emotion. An additional set of AOIs were defined for the whole image to detect when participants direct their attention to an image and to better define what the participant was looking at when or if they successfully completed the trial and were free to direct their gaze elsewhere. Separate AOIs were created for the target emotion image to differentiate between non-target images and the target image for both tasks (i.e., adult and child tasks).

Data analysis

The Tobii Eye-tracking system has a 60 Hz sampling rate and collected raw eye movement data points approximately every 16.66ms. Each data point was automatically identified with a timestamp and coordinates corresponding to gaze point locations sent to Tobii Pro Studio. Tobii Pro Studio further processed coordinates into fixations and overlayed the

coordinates onto video recording (i.e., stimuli images) for calculating eye tracking metrics that provided gaze point data for each eye, stimulus related events and AOI data, and mouse click events for caregivers. Analysis software in Tobii Pro Studio allowed the system to output descriptive statistical information for all AOIs (i.e., mean, median, mode, standard deviation, minimum, and maximum). Tobii Pro Studio software defined good data as gaze samples with usable data divided by the number of attempts. A I-VT fixation filter was applied that included a gap fill-in algorithm for data loss gaps up to 75ms and defined fixations as 150ms. Missing eye-tracking data was not replaced or interpolated beyond 75ms. All eye tracking data were automatically compiled into a single file and output by Tobii Studio with separate files for descriptive statistics also automatically calculated in Tobii Pro Studio. Output eye tracking files were preprocessed to remove extraneous space in the data file for ease of analysis. Missing clinical variables were not imputed. All subjects with missing variables were not included in the clinical variable analyses.

Eye-tracking variables assessed either fixation or visit behavior. Four fixation variables were computed and included an index of the time from trial start to initial fixation on an AOI or AOI group (i.e., time to first fixation), and index of the duration of initial fixations (i.e., first fixation duration), the total duration of fixations (i.e., FD – mean and median reflect fixation duration per trial and sum reflects total fixation duration across task), and the number of total fixations a participant made (i.e., fixation counts). Two variables were computed to assess time spent looking at media after a fixation was made including an index of the number of visits made to AOIs or AOI groups (i.e., visit counts) and an index of the total amount of time spent visiting an AOI or AOI group (i.e., visit duration). Median variables were used to avoid artifact-related

skew, as the median is more robust against variability in eye-tracking outputs (e.g., increased blinks or increased blink durations).

Several variables in both caregiver and child datasets violated normality and were transformed to a normal distribution. Variables of the same type were all transformed if one variable violated normality to ensure comparable scaling for ease of interpretation (e.g., if median time to first fixation for anger media whole image AOI violated normality all time to first fixation variables were transformed). Any transformed variables that continued to violate normality were excluded from analyses unless obvious outliers were causing the normality violation. Specifically, fixation count and visit duration analyses for both caregivers and children were conducted on a dataset with two outliers removed that severely skewed sad trial variables and mediation models with general attention bias were conducted on a dataset with one outlier removed (Table 2). Variables included in hypothesis testing were used regardless of meeting normality assumptions.

Table 2. *Detailed Report for Transformation of Data that Violated Normality*

	Normality Violation Transformation	
Child Variables	Transformed?	Transformation Type
Fixation		
Time to first fixation median	Yes	Log transformed
First fixation duration median	Yes	Log transformed
Fixation duration median	Yes	Log transformed
Visit		
Visit duration median	Yes	Log transformed
Visit count AOI analysis	Yes	Log transformed
Visit count emotion analysis	No	

Caregiver Variables		
Fixation		
Time to first fixation median	No	
First fixation duration median	Yes	Log transformed
Fixation duration median	Yes	Square root transformed
Visit		
Visit duration median	Yes	Square root transformed
Visit count	No	

Attention biases were calculated using a ratio of child visit duration (i.e., total time spent viewing images after fixating on the image) to whole image target-AOIs over neutral AOIs for all participants. All face images except the target image were neutral in the child task and were used for calculation of a general attention bias variable. A second hypothesis tests a more specific attention bias defined as a ratio of visit duration to whole image anger AOIs over neutral AOIs (i.e., anger-specific attention bias).

A composite variable was created to represent the overall caregiver emotional processing strategy. Kaiser-Meyer-Olkin Measure of Sampling Adequacy (.461) and Bartlett's Test of Sphericity (χ (6) = 4.96, p = .55) demonstrated the limited appropriateness of using a factor analysis approach to create the composite score, as each variable captured a unique amount of variance. The composite score was thus calculated using four caregiver eye-tracking variables by standardizing and summing across variables. Composite scores were computed from median target identification speed operationalized as time to first fixation on all whole image target AOIs, caregiver task accuracy operationalized as d' using mouse click data, median attention to target eye AOIs over target mouth AOIs operationalized as a ratio of fixation durations, and median total time spent viewing target whole-image AOIs operationalized using visit duration.

Statistical Analysis

Hypothesis driven correlation analyses addressed relationships between child attentional bias variables at intake, child externalizing behaviors (BASC-3), internalizing and other relevant behavior scales (e.g., BASC-3: behavior symptoms index, adaptive behaviors, developmental social disorders, etc.), prenatal substance exposure, trauma exposure, and caregiver emotion identification speed during eye-tracking. Additional correlations between eye-tracking variables and clinical variables were assessed to explore physiological relationships which were not corrected for multiple comparisons, as they were considered exploratory and hypothesis generating.

Within group and group comparisons were assessed using repeated measures ANOVAs corrected for multiple comparisons using Bonferroni corrections except for AOI analyses (N = 3) for both caregiver and child data, as well as child analyses assessing across emotions (N = 3). Fishers Least Significant Difference (LSD) was used to assess differences between levels for analyses specifically containing only 3 levels. Effect sizes were included and reported as partial eta squared (ES).

Bootstrapped conditional mediation models were conducted using model 63 in PROCESS V3.5 examining child general attention bias and externalizing problems (ECBI) to determine whether changes in emotional processing biomarkers predict changes in child externalizing behavior mediated by caregiver's facial emotional processing strategy composite variable composed of select variables supported by the emotional processing literature (caregiver face identification speed, time spent viewing target, accuracy, and attention to eyes over mouths) and moderated by trauma exposure (i.e., number of ACEs) and child age to control for inherent developmental range, (Figure 1) (Hayes, 2018). A second model was assessed using the same

originally proposed model with an attention bias variable computed using child attention to anger over attention to neutral to address a more specific relationship between attention bias and externalizing behaviors driven by attention to/from anger media (i.e., anger-specific attention bias). All continuous predictor variables were centered around zero to prevent multicollinearity among predictors and interaction items in the equation (Rose et al., 2004; Montoya et al., 2019).

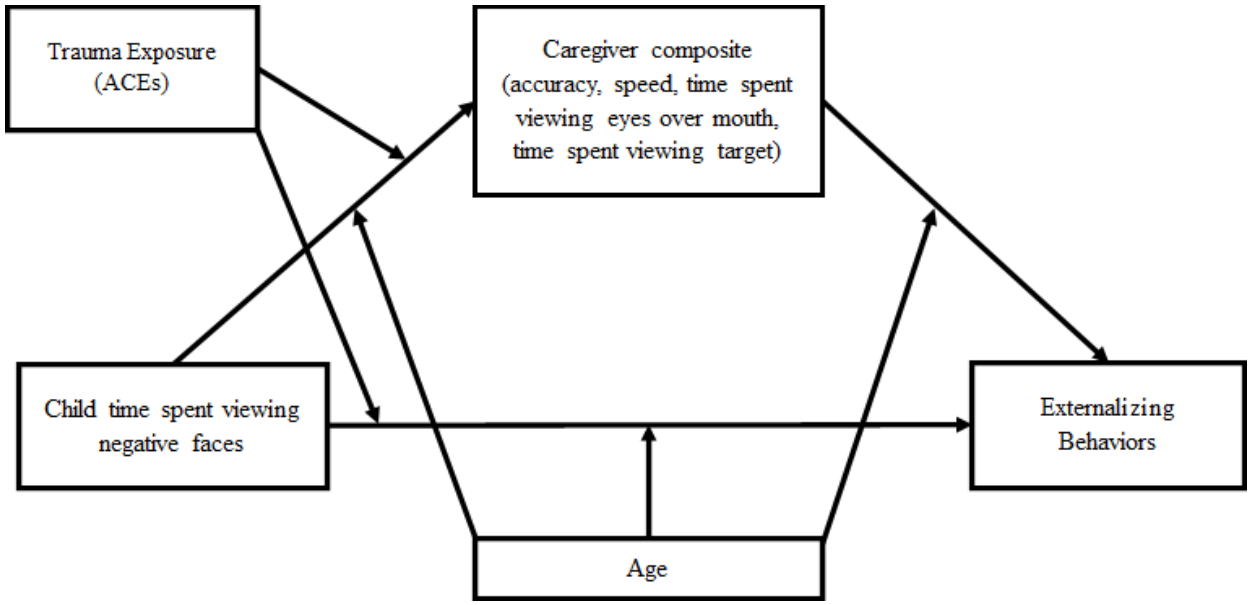

Figure 1. Mediation model for the relationship between child eye-tracking variables and child externalization behaviors.

Results

A conditional mediation model was conducted to assess a model proposing caregiver emotional processing mediated the relationship between attention biases and externalizing behaviors measured using the ECBI intensity score in children with prenatal substance exposure. The model controlled for inherent developmental range (effect of age) and the moderating influence of trauma (ACE count) on the primary relationship between attention bias and externalizing behaviors, as well as the relationship between attention bias and caregiver emotional processing. Examining the primary relationship between child attention biases and externalizing behaviors from the ECBI demonstrated a statistically significant total effect model that explained 79.68% of the variability in externalizing behaviors, $F(7, 10) = 5.60$, $p = .007$, $R^2 = .79$ (Figure 2-3; Table 3a; see appendix B, Figure B1), with a significant direct effect of child attention bias on externalizing behaviors ($\beta = -12.89$, C.I. [-25.40 -.37], $p = .045$), a significant indirect effect of caregiver composite scores ($\beta = 24.61$, C.I. [5.26 43.94], $p = .017$), a significant conditional moderating effect of age on the relationship between child attention bias and externalizing behaviors ($\beta = 11.06$, C.I. [4.54 17.52], $p = .004$), and a significant conditional interaction between caregiver composite scores and the direct moderating effect of age ($\beta = -29.85$, C.I. [-56.62 -3.08], $p = .032$) on externalizing behaviors. The conditional indirect effects of child general attention bias on externalizing behaviors were not significant suggesting no full mediation in the model (Table 3b). An unconditional interaction between the mediator (i.e., caregiver composite scores) and age was significant ($F(1, 10) = 6.18$, $p = .032$, $\Delta R^2 = .13$) but all other unconditional interactions were not significant.

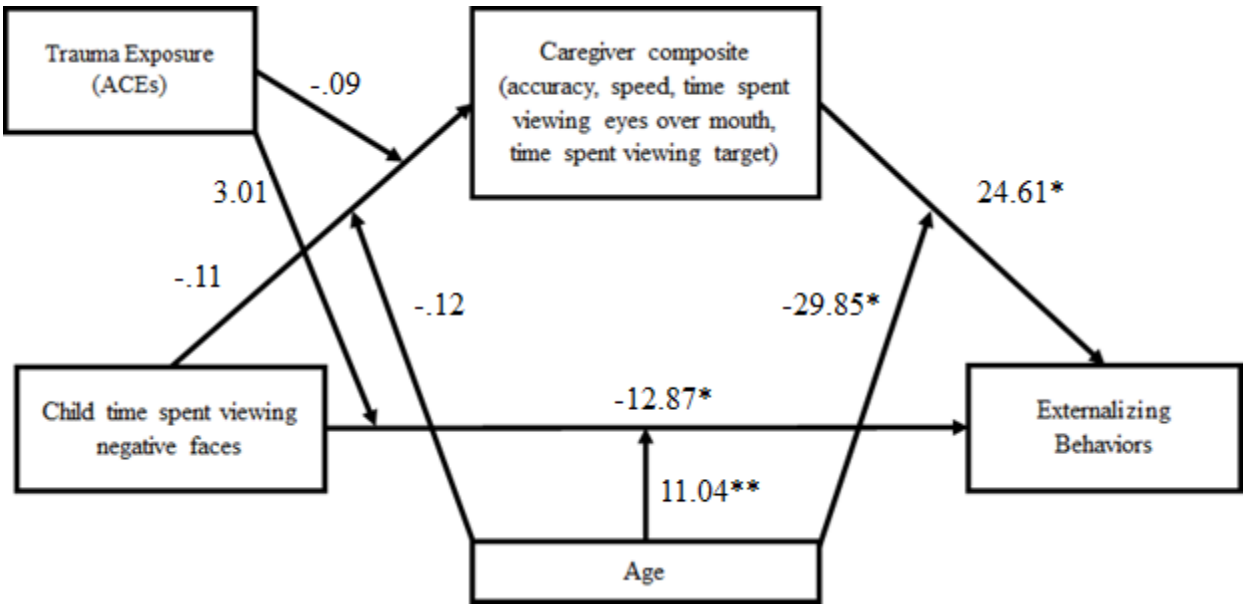

Figure 2. Mediation model and path coefficients for the relationship between child general attention bias and child externalization behaviors measured using ECBI intensity scores. Blank = N.S., *$p < .05$, ** $p < .01$, *** $p < .001$.

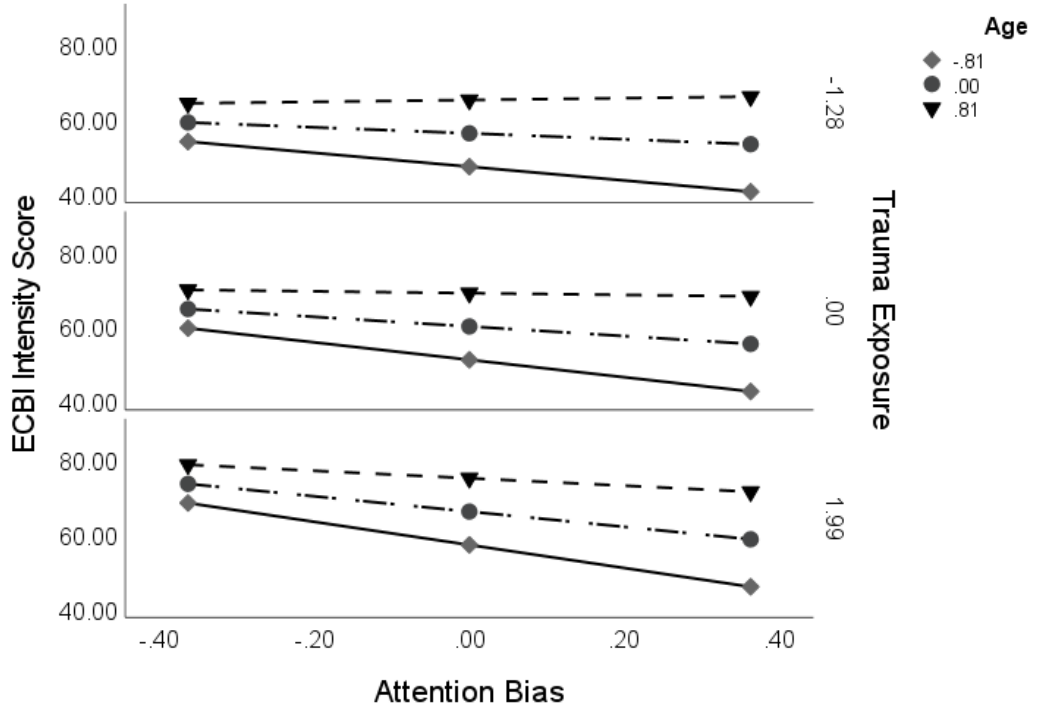

Figure 3. Data visualization for the conditional effect of the focal predictor for the general attention bias model with conditioning values at the mean and ±1 standard deviation.

Table 3a. *Conditional Direct Effects of X on Y at levels of the moderators for Primary Child Attention Bias Model using the ECBI Intensity Score*

Age	Trauma	Effect	se	t	p	LLCI	ULCI
-0.81	-1.28	-18.51	8.36	-2.21	0.05	-37.15	0.12
-0.81	.00	-23.38	9.49	-2.46	0.03	-44.52	-2.24
-0.81	1.99	-30.97	13.13	-2.36	0.04	-60.23	-1.70
.00	-1.28	-8.02	6.51	-1.23	0.25	-22.54	6.49
.00	.00	-12.89	5.62	-2.29	0.04	-25.4	-0.37
.00	1.99	-20.47	8.11	-2.53	0.03	-38.55	-2.40
0.81	-1.28	2.47	10.67	0.23	0.82	-21.32	26.25
0.81	.00	-2.4	8.49	-0.28	0.78	-21.32	16.53
0.81	1.99	-9.98	7.62	-1.31	0.22	-26.97	7.00

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Table 3b. *Conditional Indirect Effects of X on Y at levels of the moderators for Primary Child Attention Bias Model using the ECBI Intensity Score*

Age	ACE#	Effect	BootSE	BootLLCI	BootULCI
-0.81	-1.28	-9.85	46.3	-72.53	16.82
-0.81	0	-6.5	59.2	-94.01	29.36
-0.81	1.99	-1.27	112.79	-193.26	77.27
0	-1.28	0.95	13.21	-19.33	21.6
0	0	2.63	19.09	-31.8	26.36
0	1.99	5.27	48.8	-86.14	47.45
0.81	-1.28	0.11	30.74	-25.06	34.56
0.81	0	0.14	35.26	-30.46	41.24
0.81	1.99	0.18	60.16	-51.24	58.9

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

A second conditional mediation model was conducted on all dyads to assess a model proposing caregiver emotional processing as a mediator of the relationship between attention bias and externalizing behaviors in children with prenatal substance exposure measured using the externalizing score from the BASC-3. The simple regression model assessing the mediator was not significant, but there was a significant moderating effect of trauma on the relationship between child attention biases and caregiver composite scores (β = -.089, C.I. [-.15 -.03], p = .007), $F(5, 23) = 2.01$, p = .11. The total effect model assessing the primary relationship between child attention bias and externalizing behaviors using the BASC-3 externalizing score was not

significant and explained only 34.11% of the variability in externalizing behaviors, $F(7, 21) = 1.55$, $p = .20$, $R^2 = .34$ (see Appendix B, Figure B2; Table 5). The conditional indirect effects of general child attention bias on externalizing behaviors were not significant suggesting no full mediation in the model (Table 5b). No direct effect of child attention bias($\beta = -11.48$, C.I. [-30.29 7.34], $p = .22$) or indirect effect of caregiver composite scores on externalizing behaviors ($\beta = -17.02$, C.I. [-6.27 40.32], $p = .14$) were present, but a significant moderating effect of age on the relationship between child attention bias and externalizing behaviors ($\beta = 6.42$, C.I. [.32 12.52], $p = .040$) was present. There were also no significant unconditional interactions, but the test of child attention bias by mediator (i.e., caregiver composites score) interaction trended toward significance, $F(1, 20) = 4.19$, $p = .054$.

To ensure that both externalizing measures indexed similar behaviors, a one-way repeated measure ANOVA was run on z-scored ECBI and BASC-3 externalizing scores to assess differences in parent rated behavior frequency. There was a significant main effect of scale with caregivers reporting externalizing behaviors as occurring more frequently on the BASC-3 ($M = 67.44$, $SE = 11.34$) compared to the ECBI ($M = 62.50$, $SE = 11.15$), $F(1, 17) = 16.30$, $p < .001$, ES = .49.

An alternative model was run to explore the same relationship using the BASC-3 externalizing score with a subset of the sample, which included only the children that had ECBI scores (N = 18). The total effect model summary for the same model run on the dyad subset with ECBI scores was statistically significant and explained 69.49% of the variability in externalizing behaviors, $F(7, 10) = 3.25$, $p = .045$, $R^2 = .69$ (see Appendix B, Figure B3; Table 5). The conditional indirect effects of child general attention bias on externalizing behaviors were not significant suggesting no full mediation in the model (Table 5b). The total effect model

demonstrated no significant direct effect of child attention bias on externalizing behaviors (β = -8.19, C.I. [-23.79 7.41], p = .27) but a significant indirect effect of caregiver composite scores (β = 25.12, C.I. [1.02 49.22], p = .043), a significant conditional moderating effect of age on the relationship between child attention bias and externalizing behaviors (β = 9.09, C.I. [.99 17.18], p = .031), and a trending conditional moderating effect of trauma on the relationship between child attention bias and externalizing behaviors (β = 4.23, C.I. [-.39 8.86], p = .069). No significant unconditional interactions were found. Findings from the second model with the dyad subset with ECBI scores demonstrated similar patterns to the conditional mediation output using ECBI intensity scores as the externalizing behaviors measure.

A second, more specific hypothesis was tested using a conditional mediation model probing the relationship between externalizing behavior and child attention bias calculated using only anger media. The relationship between child anger-specific attention bias and externalizing behaviors appeared curvilinear when testing for a quadratic relationship where the relationship between the anger-specific attention bias and ECBI intensity scores trended toward significance (F(2, 15) = 3.43, p = .059, R^2 = .31) and the relationship between anger-specific attention bias and BASC-3 externalizing scores was significant (F(2, 27) = 4.11, p = .028, R^2 = .23). For the general child attention bias, the linear and the quadratic relationships between child attention bias and externalizing behaviors using both ECBI intensity and BASC-3 externalizing scores were not significant (Linear: ECBI: F(1, 16) = .23, p = .64, R^2 = .01.; BASC-3 Ext: F(1, 27) = .28, p = .59, R^2 = .01), but the quadratic model exhibited better fit compared to the linear model with the relationship between child attention bias and externalizing behaviors measured using ECBI intensity scores was trending toward significance (Quadratic: F(2, 15) = 3.63, p = .052, R^2 = .33.; BASC-3 Ext: F(2, 26) = 1.89, p = .17, R^2 = .13).

The model using child anger-specific attention biases demonstrated a significant total effect model that explained 84.25% of the variability in externalizing behaviors, $F(7, 10) = 7.64$, $p = .002$, $R^2 = .84$ (Figure 4-5; Table 4a; see Appendix C, Figure C1). The conditional indirect effects of child anger-specific attention bias on externalizing behaviors were not significant suggesting no full mediation in the model (Table 4b). The model exhibited several significant and trending relationships between anger-specific attention bias and externalizing behaviors measured using ECBI intensity scores. The conditional moderating effect of age on externalizing behaviors was significant ($\beta = 10.12$, C.I. [4.52 15.72], $p = .002$), the indirect effect of caregiver composite scores on externalizing behavior was significant ($\beta = 19.14$, C.I. [2.65 35.64], $p = .027$), the direct effect of anger-specific attention bias on externalizing behavior was significant ($\beta = -25.57$, C.I. [-42.92 -8.22] , $p = .008$), the conditional moderating effect of trauma exposure on the relationship between anger-specific attention bias and externalizing behaviors trended toward significance ($\beta = 2.95$, C.I. [-.24 6.13], $p = .066$), and the conditional effect of the interaction between caregiver composite scores and age was significant ($\beta = -25.08$, C.I. [-49.42 -.74], $p = .045$). An unconditional interaction between the mediator (i.e., caregiver composite scores) and one of the moderator variables (i.e., age) was significant ($F(7, 10) = 5.27$, $p = .045$, $\Delta R^2 = .08$) and the unconditional interaction between child anger-specific attention bias and age trended toward significance ($F(7, 10) = 3.88$, $p = .077$, $\Delta R^2 = .01$) but all other unconditional interactions were not significant.

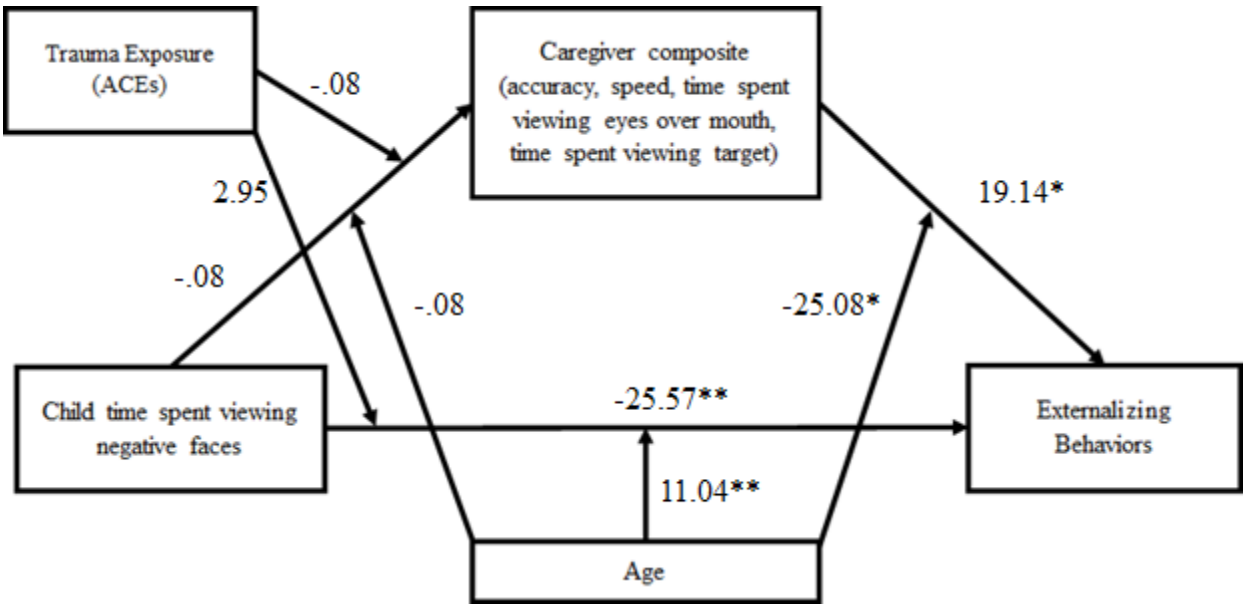

Figure 4. Mediation model and path coefficients for the relationship between child anger-specific attention bias and child externalization behaviors measured using ECBI intensity scores. Blank = N.S., *p < .05, ** p < .01, *** p < .001.

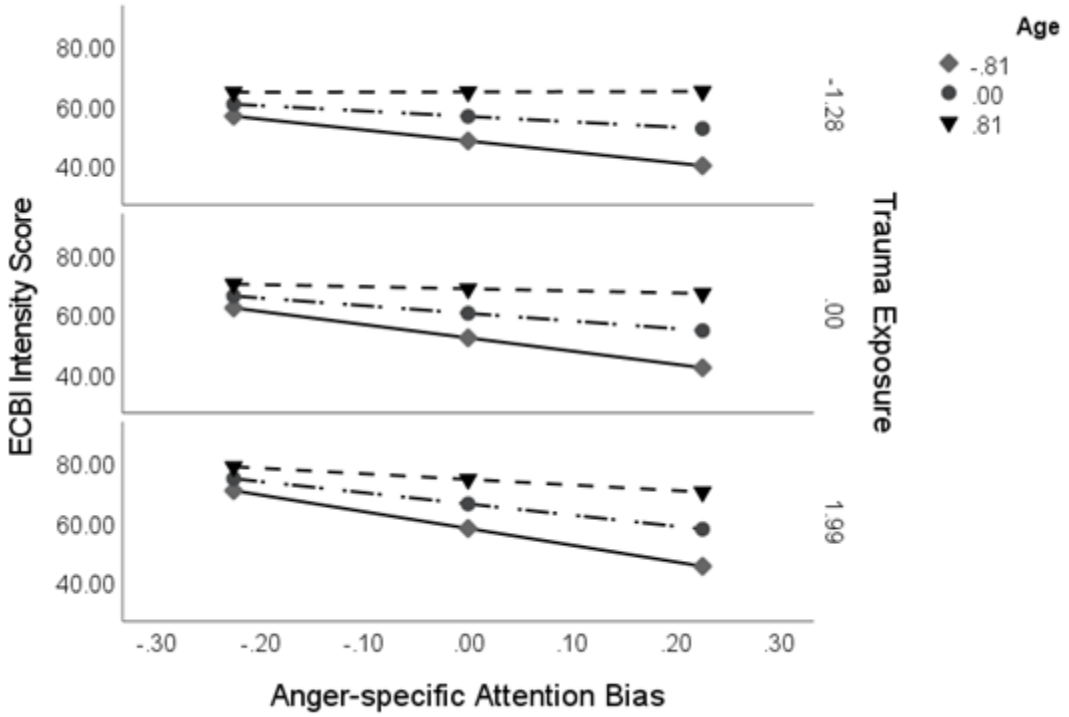

Figure 5. Data visualization for the conditional effect of the focal predictor for the anger-specific attention bias model with conditioning values at the mean and ±1 standard deviation.

Table 4a. *Conditional Direct Effects of X on Y at levels of the moderators for Child Anger-specific Attention Bias Model using the ECBI score*

Age	Trauma	Effect	SE	t	p	LLCI	ULCI
-0.81	-1.28	-36.67	11.94	-3.07	0.01	-63.28	-10.05
-0.81	0	-44.2	13.74	-3.22	0.01	-74.81	-13.58
-0.81	1.99	-55.95	19.34	-2.89	0.02	-99.06	-12.84
0	-1.28	-18.04	8.66	-2.08	0.06	-37.33	1.25
0	0	-25.57	7.78	-3.28	0.01	-42.92	-8.22
0	1.99	-37.32	12.31	-3.03	0.01	-64.77	-9.88
0.81	-1.28	0.59	13.65	0.04	0.97	-29.83	31.01
0.81	0	-6.94	10.56	-0.66	0.53	-30.48	16.6
0.81	1.99	-18.69	10.4	-1.8	0.1	-41.88	4.49

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Table 4b. *Conditional Indirect Effects of X on Y at levels of the moderators for Child Anger-specific Attention Bias Model using the ECBI score*

Age	ACE#	Effect	BootSE	BootLLCI	BootULCI
-0.81	-1.28	-20.24	303.67	-153.25	21.79
-0.81	0	-12.97	405.83	-180.45	59.75
-0.81	1.99	-1.62	882	-279.05	148.67
0	-1.28	-5.03	37.57	-49.19	20.24
0	0	-1.5	210.66	-65.08	40.22
0	1.99	4	535.19	-140.95	110.7
0.81	-1.28	0.02	58.12	-62.47	34.75
0.81	0	-0.21	294.63	-72.76	51.45
0.81	1.99	-0.55	767.26	-135.75	128.9

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Following the same methods for the primary mediation models, the initial proposed model was run using the BASC-3 externalizing score to confirm the relationship between anger-specific attention bias and externalizing behaviors. With externalizing behaviors as the primary outcome variable, the total effect model was not significant, explaining only 34.19% of the variability in externalizing behaviors, $F(7,22) = 1.63$, $p = .18$, $R^2 = .34$ (see Appendix C, Figure C2; Table 5). The conditional indirect effects of child anger-specific attention bias on externalizing behaviors were not significant suggesting no full mediation in the model (Table 5c). There was no direct effect of anger-specific attention bias (β = -15.55, C.I.[-46.28 15.17], *p*

= .31) or indirect effect of caregiver composite scores (β = 14.49, C.I. [-8.39 37.37], p = .20) on externalizing behaviors and no significant unconditional interactions. The model assessing relationships between child anger-specific attention bias and caregiver composite scores (i.e., the mediator) via simple regression was trending toward significance but explained only 32.22% of the variability in caregiver composite scores, $F(5, 24) = 1.63$, p = .079, R^2 = .32. The conditional moderating effect of trauma exposure on the relationship between child anger-specific attention bias and caregiver composite scores was significant (β = -.086, C.I. [-.147, -.025], p = .007), and the interaction between anger-specific attention bias and trauma exposure trended toward significance (β = .20, C.I. [-.017 .43], p = .069). The unconditional interaction between anger-specific attention bias and trauma also trended toward significance, F (1, 24) = 3.64, p =.069, ΔR^2 = .10.

Assessing the potential dampening effect of dyads without ECBI scores demonstrated a significant total effect model that explained 72.28% of the variability in externalizing behaviors using BASC-3 externalizing scores as the outcome variable, $F(7,10) = 3.73$, p = .030, R^2 = .72 (see Appendix C, Figure C3; Table 5). The conditional indirect effects of child anger-specific attention bias on externalizing behaviors were not significant suggesting no full mediation in the model (Table 5c). The direct effect of anger-specific attention bias on externalizing behaviors was not significant (β = 19.69, C.I. [-43.11 3.71], p = .090), but the conditional moderating effect of trauma exposure on the relationship between child anger-specific attention bias and externalizing behaviors was significant (β = 4.29, C.I. [.002 8.59], p = .049), the conditional moderating effect of age on the relationship between child anger-specific attention bias and externalizing behaviors was significant (β = 8.47, C.I. [.92 16.03], p = .031), and the indirect effect of caregiver composite scores on externalizing behaviors trended toward significance (β =

21.78, C.I. [-.48 44.04], p = .054). A simple regression model assessed the relationship between child anger-specific attention bias and caregiver composite scores (i.e., the mediator) and found the relationship was not significant, unlike the model run with the full sample, where the model explained 39.85% of the variability in caregiver composite scores, $F(5, 12) = 1.59$, p = .24, R^2 = .39. There were no significant unconditional interactions.

Table 5. *Mediation Model Path Coefficients for Models using BASC-3 Externalizing Scores as the Externalizing Behavior Variable.*

Model Path	Standardized path coefficients	
	BASC-3 Ext All dyads	**BASC-3 Ext Dyad subset (N = 18)**
General Attention Bias		
AB → CCS	.09	.11
Age → CCS	-.08	-.12
ACEs → CCS	-.09**	-.09
AB x Age→ CCS	.03	.29
AB x ACEs → CCS	.09	.05
CCS → Externalizing behaviors	17.02	25.12*
AB → Externalizing behaviors	-11.48	-8.19
Age → Externalizing behaviors	6.42*	9.09*
ACEs → Externalizing behaviors	.95	4.23
AB x Age → Externalizing behaviors	7.88	15.96
CCS x Age → Externalizing behaviors	-10.74	-23.86
AB x ACEs → Externalizing behaviors	.08	-6.38
Anger-specific Attention Bias		
AB → CCS	.09	-.08
Age → CCS	-.06	-.09
ACEs → CCS	-.09**	-.08
AB x Age→ CCS	-.08	.31
AB x ACEs → CCS	.20	.14
CCS → Externalizing behaviors	14.49	21.78
AB → Externalizing behaviors	-15.55	-19.69
Age → Externalizing behaviors	5.92*	8.47*
ACEs → Externalizing behaviors	.81	4.29*
AB x Age → Externalizing behaviors	13.38	27.23
CCS x Age → Externalizing behaviors	-9.60	-18.47
AB x ACEs → Externalizing behaviors	.72	-10.31

Note. *$p < 0.05$, **$p < 0.01$, ***$p < 0.001$, all two tailed. Mediation models all follow the same model structure as those in **Figure 1**. Abbreviations: AB = attention bias, CCS = caregiver composite score**.**

Table 5b. *Conditional Indirect Effects of X on Y at levels of the moderators for Child General Attention Bias Model using the ECBI score*

Child General Attention Bias

Age	ACE#	Effect	Boot SE	Boot LLCI	Boot ULCI
BASC-3 Externalizing All					
-0.9335	-1.5172	-2.2208	10.6669	-31.6722	10.8172
-0.9335	0	1.489	11.3435	-30.6307	15.3244
-0.9335	1.8052	5.9028	18.2454	-44.6713	27.936
0	-1.5172	-0.851	4.2718	-12.3517	5.8019
0	0	1.4837	4.4063	-10.503	6.0625
0	1.8052	4.2615	9.3905	-21.0496	13.8911
0.9335	-1.5172	-0.1251	8.0684	-15.8893	14.9329
0.9335	0	0.8345	7.0666	-13.6617	11.0663
0.9335	1.8052	1.9763	10.415	-19.4749	18.0922
BASC-3 Externalizing Subset					
-0.8112	-1.2778	-8.9704	64.2738	-73.8222	21.2603
-0.8112	0	-5.9198	71.7958	-92.644	28.4637
-0.8112	1.9943	-1.1585	147.3786	-185.374	64.8269
0	-1.2778	0.9653	63.7793	-22.7999	20.5602
0	0	2.6884	53.267	-35.3762	20.7459
0	1.9943	5.3777	83.4916	-92.7589	34.5141
0.8112	-1.2778	1.6063	69.2244	-35.3313	34.482
0.8112	0	2.0019	70.9198	-45.8247	31.7263
0.8112	1.9943	2.6193	109.5911	-77.5088	42.9269
-0.8112	-1.2778	-8.9704	64.2738	-73.8222	21.2603

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Table 5c. *Conditional Indirect Effects of X on Y at levels of the moderators for Child General Attention Bias Model using the ECBI score*

Child Anger-Specific Attention Bias

Age	ACE#	Effect	Boot SE	Boot LLCI	Boot ULCI
BASC-3 Externalizing All					
-0.92	-1.57	-3.61	12.82	-37.19	15.68
-0.92	0	3.86	14.89	-33.45	28.51
-0.92	1.79	12.4	24.78	-48.19	53.32
0	-1.57	-3.31	6.23	-19.51	6.04
0	0	1.33	5.7	-14.41	9.52
0	1.79	6.64	11.53	-22.85	23.73
0.92	-1.57	-1.71	15.29	-24.15	37.34
0.92	0	0.1	11.97	-19.1	28.22
0.92	1.79	2.18	17.6	-32.64	37.64
BASC-3 Externalizing Subset					
-0.81	-1.28	-18.84	396.36	-199.19	28.77
-0.81	0	-12.07	438.15	-224.06	66.85
-0.81	1.99	-1.51	546.97	-346.35	154.3
0	-1.28	-5.72	66.38	-57.49	21.91
0	0	-1.71	72.15	-93.35	36.32
0	1.99	4.55	130.58	-196.64	101.67
0.81	-1.28	-0.09	111.69	-68.9	50.5
0.81	0	1.17	93.77	-98.84	52.91
0.81	1.99	3.12	189.89	-199.6	121.07
-0.81	-1.28	-18.84	396.36	-199.19	28.77

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Clinical Correlations

Clinical correlations of note included those that support primary hypotheses and those that contribute to mediation model interpretations (Figure 6). The number of trauma exposure types was significantly correlated with the BASC-3 developmental social disorders subscale, an index of behaviors typically noted in autistic children. Child ECBI intensity (i.e., frequency) scores were significantly correlated with all BASC-3 composite scores except emotional self-control suggesting that the ECBI does not index emotional regulation. Child ECBI problem scores were only correlated with BASC-3 externalizing and negative emotionality, suggesting

that ECBI problem scores may reflect a more specific index of externalizing behaviors compared to intensity scores.

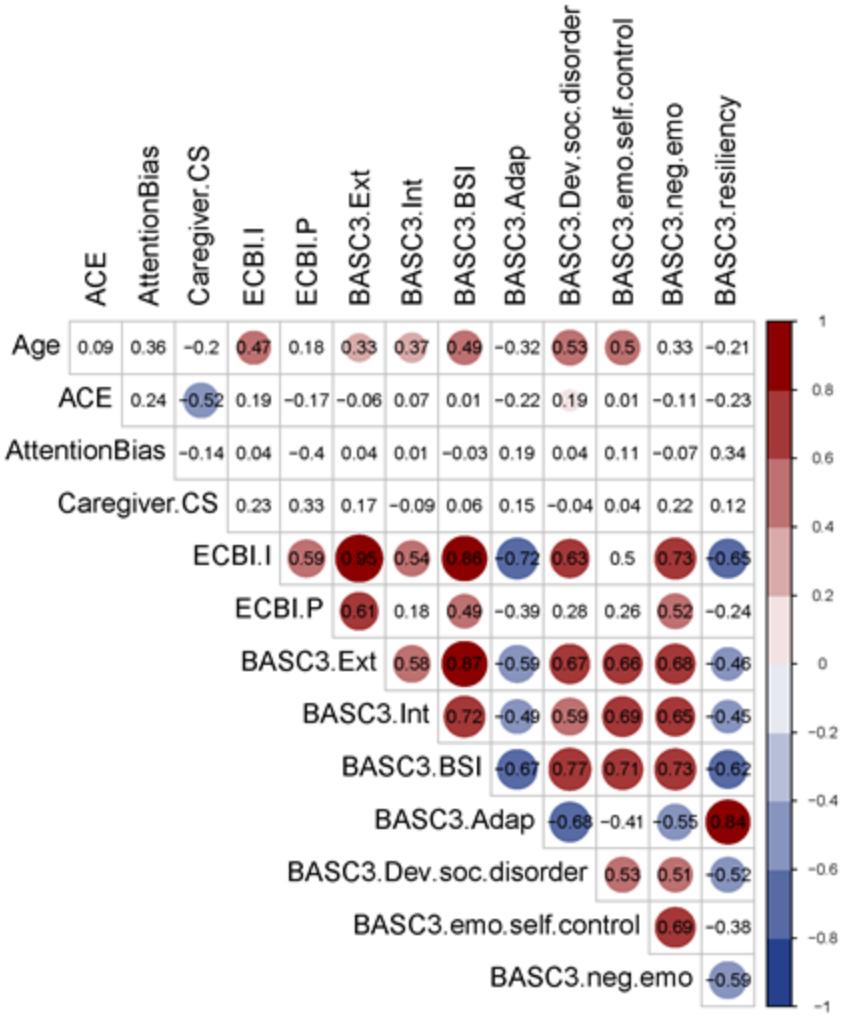

Figure 6. All correlations are Pearson's rho. All significant correlations are listed on colored circles. Blank = N.S.

Full scale IQ and most clinical measures of cognitive function did not share relationships with trauma exposure, child attention bis, caregiver composite scores, or behavior measures on the BASC-3 (Figure 7). The fluid reasoning (FRI) and processing speed indices (PSI) from the WPPSI-IV demonstrated significant direct relationships with ECBI intensity scores and the FLI also exhibited a significant direct effect with child attention bias. Assessing relationships

between child trauma exposure and caregiver variables that went into the composite score showed an inverse relationship between trauma exposure and caregiver time spent viewing the target, rho = -.48, p = .025. Caregivers generally spent less time viewing target media when children had increased trauma exposure.

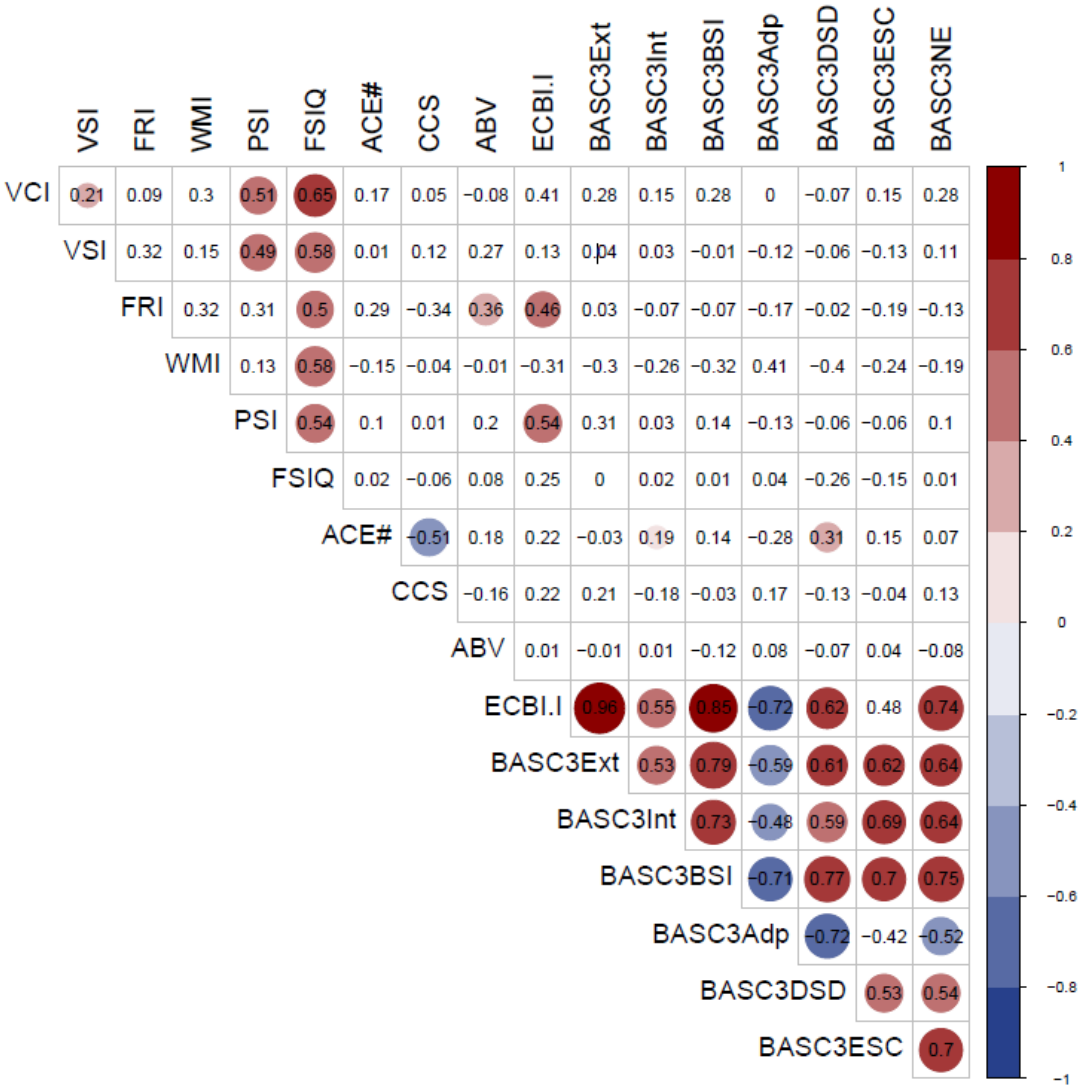

Figure 7. All correlations are Pearson's rho. All significant correlations are listed on colored circles. Blank = N.S.

Child Exploratory Outcomes

To assess whether model outcomes were associated with alcohol or substance related to oculomotor abnormalities, we ran two independent samples t-tests to assess ECBI intensity and BASC-3 externalizing score by confirmed or suspected alcohol exposure (i.e., combination of confirmed and suspected). Children in the sample with confirmed or suspected alcohol exposure were not significantly difference from children without exposure on either ECBI intensity (t(16) = -.39, p = .70) or BASC-3 externalizing scores (t(27) = -.40, p = .69).

A 2x3 repeated measure ANOVA was run to assess target AOI (eye and mouth AOIs) by emotion (anger, fear, and sad) effects for median visit duration (VD). Analysis of mean VD showed a main effect of AOI where children attended longer to mouth AOIs (M = -.38, SE = .02) over eye AOIs (M = -.47, SE = .03), F (2, 21) = 5.64, p = .027, ES = .20.

A one-way repeated measure ANOVA was run to assess the total number of visits to whole image AOIs across emotions and found no main effect of emotion, F(2, 25) = 2.27, p = .12, ES = .15. Age was a significant covariate for the main effect of emotion, suggesting age influenced the number of times children visited media expressing negative emotions, F(1, 28) = 10.79, p = .003. An alternative analysis was conducted to include neutral images where neutral was computed by dividing the total number of visits to neutral images by the number of neutral images displayed within a trial (i.e., 5) and found a significant main effect of emotion, F(2, 24) = 3.56, p = .029, ES = .31. The age covariate trended toward significance (F(1, 26) = 3.72, p = .065) and the interaction between emotion and age was significant (F(3, 24) = 3.35, p = .036, ES = .29), supporting the conclusion that age may influence the number of times children visited certain media types. Age shared a direct relationship with visit count for all three emotions (anger: rho = .47, p = .009; Fear: rho = .49, p = .007; Sad: rho = .57, p =.001). Differences across

emotions were not significant after correction for multiple comparisons but demonstrated a pattern of decreased time spent attending to sad media and more time spent attending to fear (neutral and anger appeared equal).

One-way repeated measure ANOVAs were run to assess median time to first fixation (TFF) for whole image target AOIs across emotions with no significant main effect of emotion, $F(2, 28) = .44$, $p = .65$, ES = .03. Initial fixation patterns were explored used two one-way repeated measure ANOVAs to assess median first fixation duration (FFD) across emotions. The durations of initial fixations were not significant suggesting initial fixations were similar across all emotions, ($F(2, 28) = .56$, $p = .58$, ES = .04. A two-way repeated measure ANOVA assessed median fixation duration (FD) across emotions by AOI for target media and found no significant main effects of emotion ($F(2,20) = 1.60$, $p = .23$, ES = .14) or AOI ($F(2,20) = 1.43$, $p = .26$, ES = .13) and no significant interaction between AOI and emotion ($F(2,20) = .22$, $p = .93$, ES = .05).

Caregiver Exploratory Outcomes

Two one-way repeated measure ANOVAs were run to assess median VD and the total number of visits made to media across emotions. Assessing VD across emotions resulted in a significant main effect of emotion, $F(5,25) = 3.36$, $p = .019$, ES = .40. There were no significant differences between emotions after correcting for multiple comparisons. Testing for differences across emotions for the number of visits made by caregivers with neutral included in the analysis resulted in a significant main effect of emotion, $F(5,25) = 23.11$, $p > .001$, ES = .82. Caregivers made significantly more visits to sad media ($M = 26.27$, $SE = 1.25$) compared to all other emotions (Disgust: $M = 22.90$, $SE = 1.02$; Fear: $M = 20.77$, $SE = 1.07$; Happy: $M = 22.67$, $SE = 1.13$; Neutral $M = 21.13$, $SE = 1.00$), except anger media ($M = 25.77$, $SE = 1.14$). Caregivers seemingly visited fear the least, but significantly differed from anger, sad, and happy. Overall,

caregivers appeared to attend more strongly to anger and sad compared to other emotions. The neutral variable violated normality and therefore a second analysis was conducted to assess relationships without neutral included. Patterns were comparable to findings when neutral was included, $F(4, 26) = 28.45$, $p > .001$, ES = .81. Sad ($M = 26.27$, $SE = 1.25$) and anger ($M = 25.77$, $SE = 1.14$) were visited more frequently compared to other emotions (Disgust: $M = 22.90$, $SE = 1.02$; Fear: $M = 20.77$, $SE = 1.07$; Happy: $M = 22.67$, $SE = 1.13$).

A one-way repeated measure ANOVA was run to assess median TFF for whole image AOIs across emotions. Analysis of median TFF found a significant main effect of emotion, $F(5, 25) = 7.04$, $p > .001$, ES = .59. Caregivers were significantly slower to fixate on neutral media ($M = 1.67$, $SE = .06$) compared to anger ($M = 1.45$, $SE = .05$) and happy ($M = 1.33$, $SE = .05$) media. TFF was significantly faster for happy media compared to fear ($M = 1.47$, $SE = .06$) and sad ($M = 1.65$, $SE = .06$) media, in addition to neutral. Disgust was not significantly different from all other emotions but slotted closely to other negatively valenced emotional stimuli ($M = 1.48$, $SE = .05$). Finally, caregivers were significantly slower to fixate on fear media compared to anger media. Caregivers demonstrated patterns of increased TFF for all emotions compared to neutral, except in the case of median TFF for fear media which was comparable to TFF for neutral media. To further explore initial fixation patterns, a one-way repeated measure ANOVA was run to assess FFD across emotions. The durations of median initial fixations were not significant suggesting initial fixations were similar across all emotions, $F(5, 25) = .24$, $p = .95$, ES = .04.

Two one-way repeated measure ANOVAs were run to assess total FD (i.e., sum) across emotions and target AOIs. FD across emotions did not significantly differ for caregivers. The assessment of FD across target AOIs showed a significant main effect of AOI, $F(2,28) = 7.04$, p

> .001, ES = .59. Total FD was significantly different across all AOIs with caregivers attending to whole image AOIs longer (M = 8.97, SE = .26) than mouth (M = 7.59, SE = .36) or eye (M = 3.70, SE = .29) AOIs, though whole image AOIs contain both eye and mouth regions and therefore background viewing was difficult to discern from summation of facial region viewing and time spent moving toward the face. Caregivers viewed mouth AOIs for significantly longer durations compared to eye AOIs, similar to patterns noted among children. The number of fixations (i.e., fixation counts) caregivers made across emotions were assessed using a one-way repeated measure ANOVA and found no significant differences across emotions, $F(2,20) = .73$, $p = .61$, ES = .14.

Discussion

Child maltreatment is a global public health concern with long term negative consequences for social, emotional, and cognitive wellbeing (Merrick et al., 2018; Smith & Pollak, 2020). Clinical populations with PSE reflect the more extreme end of trauma exposure and addressing engagement with emotionally valanced stimuli via biomarkers help to define physiological mechanisms of emotional processing disruption more concretely. Early outcomes of populations with PSE tend to reflect increased externalizing and internalizing symptoms compared to children with reduced numbers of ACEs/adversity exposures increasing the need for physiological indexes of underlying disruptions (De Bellis et al., 1997). However, most approaches to assessing consequences of child adversity exposure focus primarily on child outcomes without including caregivers as a developmental context. Results demonstrate physiologically characterized emotional literacy in the caregiver is relevant to the relationship between negative emotion processing in children with PSE and ACEs and child behavior. Current findings suggest caregivers create a behavioral environment that influences child

behavior by mediating the relationship between physiological emotional processing mechanisms and externalizing behavior problems.

Primary Mediation Results

Overall model results suggested caregiver emotional processing may partially mediate the relationship between child attention biases and externalizing behaviors indicating caregiver response/attention patterns to emotionally valenced stimuli plays a key role in externalizing symptoms of internal emotional processing disruptions. Significant paths within models indicated variables have an impact on externalizing behavior, but full mediation of the relationship between child attention bias and externalizing behaviors was underpowered while including and considering moderators, though results may reflect partial mediation. Results demonstrate caregiver influence on child outcomes related to emotional processing with current results largely underpowered to explore full mediation of the proposed model. Significance for ECBI intensity scores as the primary model output variable demonstrates the influence of caregivers on negative shifts in child behavior toward problematic behavioral profiles when the child exhibits valence-specific emotion attention biases. While the current model does not assess bidirectionality, relationships noted within current models are potentially bidirectional in nature. Bidirectional relationships between caregiver emotional processing and both child attention biases and externalizing behavior may impact physiology such that bidirectional physiological responses mechanistically underlie caregiver-child interactions and long-term outcomes associated with negative behavior profiles (Atzaba-Poria et al., 2017). Modeling the moderating influence of age and trauma provides insight into physiological shifts contributing to behavior as a product of both developmental trajectory and adversity exposure (Rose et al., 2004). Findings support the conclusion that caregivers are an important developmental context for early

emotional processing development, specifically in young children between the ages of 3 and 7 (Cervin et al., 2021; Wolicki et al., 2021). As the developmental range of the current study was larger compared to other studies assessing attention bias as a biomarker of adversity exposure, we also conclude caregivers are important emotional processing mediators during trauma-specific sensitive periods (Davies et al., 2018; Davies et al., 2020a; Stevens et al., 2018).

Limited dyads within the sample had ECBI scores, prompting the use of the BASC-3 externalizing score to assess the same relationship. Interestingly, while mediation models were generally significant using the subset of individuals with ECBI scores, models using the entire sample with BASC-3 scores as the externalizing measure were not. Dissimilarity in model findings reflects an interesting conundrum, as we would expect comparable model outputs between both measures because both externalizing scales index similar behaviors. BASC-3 externalizing results raised questions regarding the external validity of the ECBI model results prompting further evaluation of the relationship between BASC-3 externalizing scores compared to ECBI intensity scores. The BASC-3 externalizing score and the ECBI intensity score were significantly different with caregivers reporting more frequent externalizing behavior problems on the BASC-3 compared to ECBI, even when excluding cases pairwise. Caregivers either reported different behavior frequencies between measures, or the measure scaling prompted responses that captured behaviors differently. As parents are generally stable in assessing these types of behaviors across time (Lecavalier et al., 2006), the second interpretation is the more likely one. Importantly, the administration of the ECBI was not randomly missing across the sample but was selectively administered in the clinical setting only when caregivers reported significantly problematic externalizing behaviors during any portion of the intake interview.

As models using ECBI and BASC-3 externalizing scores as indices of externalizing behavior resulted in dissimilar outcomes, we explored whether children without notable externalizing behavior problems dampened model effects when assessing the whole sample on BASC-3 externalizing scores. Models run using BASC-3 externalizing scores on the subset of dyads with ECBI scores were significant suggesting the modeled relationships primarily exist for children with significant problem behaviors. We deduced that children in the sample with behavior profiles in the normal range dampened the total model effect of models testing the relationship between attention biases and externalizing behaviors when included with children who exhibit clinically significant externalizing behavior problems, as evidenced by the necessity of the ECBI. Findings demonstrate the emergence of two subpopulations generated by using ECBI as a proxy for thresholding where modeled relationships primarily exist for those with significant externalizing behavior problems (Wiggins et al., 2015).

Identification of a potential subpopulation is not surprising, given the difficulty disentangling genetic, epigenetic, and prenatal/postnatal familial environmental effects on child development and later behavioral profiles (Knopik et al., 2019). PSE uniquely exerts environmental influences on later development with long term impacts on behavior and related behavioral outcomes and added adversity exposure further complicates underlying disruptions (Knopik et al., 2019; O'Brien & Hill, 2014; Richardson et al., 2013; Sonon et al., 2015). The role of the caregiver in emotional processing of children reflects a potential developmental risk when reduced emotional processing capacity of the caregiver feeds into the relationship between child physiology and behavior. Disruptions to caregiver-child relationships or maladapted dynamics put children at increased risk for both chronic and toxic stress which negatively impacts emotional development (Bethell et al., 2017). Alternatively, results may reflect physiological

shifts related to certain types of substance exposures that impact oculomotor control (e.g., alcohol and opioids) (Lambert & Peeler, 2019). Reduced caregiver emotional literacy may account for some variability resulting in the presence of a subpopulation, but the current results reflect a level of variability within populations with PSE, such that some factor beyond substance exposure and trauma influences the relationship between child attention biases and externalizing behaviors (Smith & Pollak, 2020; Wiggins et al., 2015).

Anger Specific Attention Biases

Reducing the child attention bias variable to include only anger media and running the same proposed model demonstrated minorly superior performance to models with the media non-specific attention bias variable, but generally performed similarly. More direct effects were trending or became significant when the model was reduced, suggesting anger potentially drives outcomes noted in the general child attention bias model. While driving mechanisms remain fully unclear, anger-specific attention bias findings support model simplification. Interestingly, the relationship between child anger-specific attention bias and externalizing behaviors appears curvilinear. Though several models were significant, certain model effects may reflect the nonlinear nature of attention biases, of which anger-specific models demonstrates a clearer relationship (Hayes, 2017). Children higher in externalizing behaviors appear to exhibit both increased and decreased attention to negative emotions, for which anger shows a stronger effect. Varying patterns of attention captured with the current sample potentially reflects varying emotional processing alterations produced by different types of adversity exposure. Abuse tends to produce sensitization responses where children are more likely to attend to, or categorize, facial expressions as angry and attend more strongly to negative emotions (Schackman & Pollak, 2014; Smith & Pollak, 2020; Davies et al., 2020a). Other types of maltreatment (e.g., neglect)

produce delays in processing and perceiving emotions with increased rumination mediating the relationship between other maltreatment types and attention biases toward sad facial expressions (Romens & Pollak, 2012; Smith & Pollak, 2020). Household disruptions, or nonstable home environments, may result in similar attention pattern outcomes to child maltreatment (i.e., interparental conflict results in both attention biases toward and away from negatively valenced emotions, where anger showed the strongest effect) (Davies et al., 2018, 2020a, 2020b).

Across all models, the conditional effect of the focal predictor demonstrated a relationship showing increased externalizing behaviors for those who exhibited general attention biases away from negatively valenced emotional stimuli and less externalizing behaviors for those who exhibited attention biases toward negatively valenced emotional stimuli (see Appendix B; Figure 3, 5). The split attention bias effect decreased with increasing age, regardless of trauma exposure suggesting age impacts the strength of the relationship between attention biases and externalizing behaviors, potentially due to the addition of more sources of environmental influence as children age. However, those with increased trauma exposure generally exhibited increased externalizing behaviors where children exhibiting attention biases away from negatively valenced stimuli demonstrating the greatest effect. Generally, age effects may reflect accumulation of ACEs because adversity accumulation demonstrates a graded relationship with behavior problems in childhood and into adolescence with intermittent adversity exposure playing a greater role in the development of negative behavior profiles (Evans et al., 2013; Schroeder et al., 2021).

Primary Correlations and Clinical Outcomes

Correlation outcomes were limited to direct relationships between clinical measures on the BASC-3 and ECBI intensity scores and the inverse relationship between caregiver composite

scores and trauma (i.e., ACEs). Age correlations suggest children exhibit increased externalizing and internalizing symptoms with increases in age. Age was significantly correlated with the BASC-3 developmental social disorder content scale suggesting internalizing, externalizing, and emotion regulation difficulties may reflect developmental-related difficulties. Caregiver composite scores correspond to increased numbers of trauma exposure types, suggesting children with caregivers that exhibit reduced emotional processing capacity tend to have higher numbers of trauma exposure types. Most caregivers were not the biological parents of their child, but the correlation in combination with conditional mediation results suggests children with higher ACEs may have increased behavior problems when caregivers exhibit reduced emotional processing capacity. Further, trauma exposure (i.e., ACEs) was correlated with the developmental social disorder content scale of the BASC-3. The developmental social disorder context scale is an index of behaviors typically noted in autistic children. Increases in trauma exposure with increases in social development disorders suggests the developmental consequences of trauma exposure may shift outcomes toward behaviors typical of autistic children. Though developmental delays as a product of trauma may result in autistic traits, increased abuse is also likely directed at individuals exhibiting increased social developmental disorder symptoms because persons with autistics traits are at increased risk for interpersonal victimization (Roberts et al., 2015).

The general lack of correlations between trauma exposure, child attention bias, caregiver composite scores, and behavioral measures suggest these measures were specific to emotional regulation and social processing, and do not index broader intellectual ability or developmental delays. A notable conclusion from conditional mediation model analyses was the presence of a subpopulation of children with greater externalizing behavior problems for which the modeled

relationships best fit. The correlations between FRI and PSI and both attention bias and ECBI intensity scores suggests that children exhibiting attention biases and greater externalizing behaviors may generally thrive better in cognitive domains that require more fluid or visual reasoning and may be able to process that information at a greater speed.

Child Exploratory Outcomes

Exploratory analyses were designed to nuance the discussion on attention patterns underlying primary model outcomes and explore gaze variables. Children attended longer to mouth AOIs compared to eye AOIs suggesting a potential bias away from eyes. Cognitive modeling posits attention biases to threat reflect reduced trait anxiety/susceptibility to affective disorders and evidence supports interindividual differences (Davies et al., 2018; 2020a; 2020b; Veerapa et al., 2020). Attention biases away from eyes may reflect a coping mechanism, though it could be more useful to attend to central or lower regions of the face over eye regions when processing emotional expressions. Preferential attention or attentional biases toward faces and eye regions are widely accepted with evidence primarily supporting biases in attention toward social stimuli, not specific facial regions. More recent work supports the notion that human attention is not biased toward specific facial features, including toward eye regions (Pereira et al., 2020). Statistically significant decreases in attention to eye regions remains of particular interest because children with prenatal substance exposure exhibit increased behaviors typical of neurodevelopmental disorders (e.g., autism spectrum disorder) and autistic children exhibit fixation decreases to facial regions compared to neurotypical children (Griffin & Scherf, 2020; Kwon et al., 2018). Additional findings show autistic children and adults exhibiting decreased attention to central features of the face, including decreased attention to eyes or eye regions compared to typically developing controls (McPartland et al., 2011).

Caregiver Exploratory Outcomes

Caregivers exhibited similar fixation patterns to children with longer fixations on mouth AOIs compared to eye AOIs. Caregiver AOI fixation behavior likely reflected search strategy and general task behavior (Pereira et al., 2020). Caregivers were slower to fixate on negative emotionally valenced facial stimuli compared to positive. Interestingly, caregivers exhibited an initial fixation bias away from emotionally neutral facial stimuli suggesting a preference for emotionally valenced facial stimuli. Mixed results exist demonstrating the existence of positive and negative attention biases as measured by number of visits in a manner that may reflect age and task-dependent influences (Kauschke et al., 2019). Age is a factor that influences cognitive task performance. We observed no age effects for initial fixation behaviors with counterbalancing media type and target location correcting for any search strategy-specific behavioral biases suggesting our findings reflect an attention bias away from negatively valenced facial stimuli (i.e., a positivity bias with positive faces processed more quickly). Studies using naturalistic facial stimuli (e.g., facial stimuli from the NimStim database) more frequently conclude in favor of a positivity bias (Kauschke et al., 2019). Recent evidence supports a link between trauma exposure and attention to negatively valenced facial stimuli over neutral (Lazarov et al., 2021). While findings from TFF suggested a bias, the amount of time spent in an initial fixation was evenly distributed across all emotions and this effect was robust to artifact. Children exhibited a similar trend with TFF biases and evenly distributed fixation time observed across initial fixations. Adult negativity biases tend to show a stronger effect for anger or threating stimuli and mediation models agree, but more nuanced caregiver analyses showed attention biases away from negativity for which sad media demonstrated a stronger effect (Kauschke et al., 2019; Lazarov et al., 2021). We find that more nuanced negative emotions do

not show the same attention biases away from faces expressing negative emotions. Sadness, for example, may reflect a more complex relationship between child and caregiver emotional processing where patterns of stronger attention reflect reduced emotional processing in the caregiver.

Conclusions and Future Directions

Measuring emotional attention to valenced stimuli as a biomarker for maladjustment in caregiver-child dyads reflects a unique approach to addressing sociocognitive aspects of emotional processing and regulation in at-risk populations. Current results support complex interplay between child emotional and caregiver emotional processing with respect to child externalizing behavior. Specifically, the aforementioned effect exists for children with externalizing behavior problems suggesting additional factors may impact the extent to which early adversity exposure impacts developmental outcomes in children with PSE, with respect to behavior. Future models would benefit from insight into bidirectionality of relationships within the current models. The current models assess static relationships where dynamic influence may exist and further insight into how factors impact behavioral outcomes are necessary to fully understand the relationship between attention biases and externalizing behaviors with caregivers included as a factor. Further, future assessments should assess adaptability and resilience capacity as moderators of the relationship between caregiver emotional processing strategy and child externalizing behaviors (Fritz et al., 2018).

Establishing eye-tracker based emotional attention as a biomarker for emotional processing deepens current mechanistic understandings for efficacy of behavioral interventions addressing the emotional environment between caregivers and children to change long-term behaviors and impact caregiver-child relationships across a broad range of socio-emotional

contexts, potentially mediated by individual differences in emotional processing strategies. Future plans include assessing the efficacy of behavioral interventions like Parent-Child Interaction Therapy (PCIT). PCIT is a mechanism recommended to address disruptive behavior in children by targeting child communication and parenting behaviors including identifying and expressing emotions (Lieneman et al., 2017). PCIT reports success in both child and caregiver behavioral modification, but there remains limited demonstrable evidence to understand the underlying mechanism related to the increased capacity for emotional regulation or emotional processing (Lienemean et al., 2019). A biomarker would improve current understandings of PCIT efficacy in terms of the proposed physiological changes and would facilitate formation of novel therapeutics designed to address underlying mechanisms of aversity exposure, including populations exposed to substances prenatally, rather than behavioral symptoms.

Limitations and Assumptions

A lack of universal adversity categories to define adversity contributes to a lack of consistency in defining, measuring, and quantifying childhood adversity. Sensitivity to adversity is biologically determined and ACEs provide a nice framework to think about how external factors interact with physiology, but ACE categories are still broadly defined and not regularly used to assess adversity exposure in children during wellness exams. Further, assessments of ACEs were limited to trauma screeners and medical histories collected from caregivers during intake examinations at the Child Study Center, limiting the way we assess how physiological variables interact with quantifiable measures of adversity exposure. Current quantifications are limited to the number of ACE categories a child experiences reported to clinicians and not the overall number of total adversity exposures. The difficulty and complexity of assessing a child's

net vulnerability to adversity and the factors that contribute to negative developmental outcomes limited individual assessments of adversity on reported variable outcomes.

Externalizing behaviors were quantified from parent report measures. The ABC clinic collects two separate measures that index externalizing behaviors, the BASC-3 and the ECBI. All caregivers were asked to complete BASC-3 forms during intake, but only caregivers that reported externalizing behaviors during intake were asked to complete the ECBI form. There were significant differences between ECBI scores and BASC-3 externalizing scores suggesting 1.) caregivers were reporting behaviors differently between measures, or 2.) the measures index externalizing behaviors differently that may warrant further investigation. Further, evidence maintains that caregiver-reports and clinical observations are not interchangeable in construct comparison. Caregivers may report inflated or biased behavioral problems based on perception of child behavior though known to report problem behaviors and stress consistently over time (Lecavalier et al., 2005; Zahidi et al., 2019). Future efforts endeavor to include clinician rated behavior reports to gauge externalizing behaviors in caregiver-child interactions (e.g., Dyadic Parent-Child Interaction Coding System or other similar measures)

Attention biases to/from negatively valenced stimuli reflected potential coping mechanisms used to deal with negative social stimuli or situations. We assumed the relationship between biased attention and negatively valenced social stimuli implied a coping mechanism directly related to coping with negatively valenced social situations. Adversity feeds into a complex developmental framework that incorporates both micro and macro contexts/factors that contribute to vulnerability. Without access to more bioecological, relational, and contextual factors it was difficult to make causal links between ACE exposure and eye-tracking outcomes. The addition of pupillometry would improve physiological resolution and provide access to

information about autonomic reactivity during tasks designed to elicit natural responses to negatively valenced social stimuli. Further, the social nature of certain variables assessed will depend on social context for which outcomes may be predominately applicable to North America.

Lastly, prenatal substance exposure as a variable in the current study referred to any type of substance utilization during pregnancy. The current sample was not large enough to split by substance type to evaluate the effects of specific substance exposures on underlying emotional processing. Effects of some substances on physiological mechanisms have been proposed, including emotional processing. However, there are benefits to addressing prenatal substance exposure generally as a form of physical trauma (i.e., maltreatment) in relation to the potential ensuing effects on emotional processing. Additionally, children with PSE tend to have notable oculomotor deficits with may impact interpretation of results in those with PSE and/or diagnosed with FAS although we found no systematic distribution of children with FAS across behavior intensities (Paolozza et al., 2014).

Chapter Two

Pupillometry as a Biomarker of LC-NE Dysregulation in Children with Prenatal Substance Exposure

Keywords:

locus coeruleus-norepinephrine; trauma; child behavior problems; pupillometry

Abstract

Early Adversity exposure has a profound effect on the activity of major neuroregulatory systems resulting in long-term consequences for cognition, emotion regulation, and general health. Early life stress associated with adversity exposure influences locus coeruleus-norepinephrine (LC-NE) function resulting in hyperreactivity of the LC-NE arousal system promoting a sustained stress response. Physiologically defined markers of adversity exposure are necessary to assess mechanisms linking adversity exposure and negative life outcomes (i.e., cognitive deficits, health conditions, early mortality, etc.). Pupil diameter is a potential biomarker that is typically used to index the LC-NE system in response to emotionally valenced stimuli for gauging autonomic arousal and stress responses. Eye-tracking evaluated the relationship between pupil diameter in response to emotionally valence stimuli and 1.) caregiver pupil diameter in response to emotionally valenced stimuli and 2.) emotional processing composite scores in caregivers to determine arousal response to emotionally valenced stimuli as potential mediators. Thirty caregiver-child dyads participated in an emotional identification task using eye-tracking. Overall results demonstrated caregiver emotional processing mediation of the relationship between LC-NE reactivity predicts decreased security in the caregiver-child relationship and increased externalizing behaviors in the child, but not caregiver pupillometry. Results suggest caregivers produce and maintain a biobehavioral environment that impacts development of emotion regulation capacity in the child positing child pupillometry as a biomarker of LC-NE dysregulation.

Background

Adverse childhood experiences (ACEs) are known to impact neural function leading to increased risk for negative physical and mental health outcomes. Adversity exposure early in life can permanently modify the activity of major neuroregulatory systems resulting in long-term neurobehavioral consequences (Anda et al., 2006; Herzog & Schmahl, 2018). Adults with a history of ACEs exhibit complex clinical profiles including co-occurring mental and somatic conditions (e.g., PTSD, depression, obesity, and diabetes) making ACEs and general trauma exposure important factors to address during development (Herzog & Schmahl, 2018).

Locus Coerueus and Norepinephrine System

Early life stress and trauma affect HPA-axis reactivity and locus coeruleus-norepinephrine (LC-NE) function leading to altered stress reactivity later in life (Anda et al., 2006; De Bellis et al., 1997). Evidence from animal studies shows increased locus coeruleus (LC) activity and norepinephrine (NE) release in response to stressors with prolonged stress associated with later hyperresponsivity. Specifically, the noradrenergic system (i.e., LC-NE) plays a key role in stress with prolonged exposure to adversity/stress resulting in long-term effects on genetic expression of LC alpha-2 noradrenergic receptors. Reduction of alpha-2 receptors impacts feedback inhibition from the noradrenergic system and results in heightened LC/NE responses to later stressors (Anda et al., 2006). Sustained stress responses associated with LC-NE hyperresponsivity include chronic psychopathology (e.g., anxiety, depression, PTSD, etc.) and physical health conditions associated with increased risks for mortality (e.g., cardiovascular disease and hypertension) (Grueschow et al., 2021).

The LC projects widely throughout the brain making connections within the brainstem and with the thalamus, limbic system, and neocortex. Widespread connections from the LC are

accompanied by distribution of NE throughout the brain with neuromodulatory effects on physiological state and adaptive behavior (Morris et al., 2020). Broadly, the nature of NE secretion in terminal regions alters global neural functions, including attention and arousal. Physiological arousal affects how individuals allocate attention reflecting a mediation effect between stress and important behaviors (e.g., learning) (Whiting et al., 2021). The LC is important for threat-related learning and memory with mechanisms including both plasticity of hippocampal projections from LC and LC influence on long-term potentiation. One memory related outcome associated with connections between LC and limbic structures is the development of attentional biases (Morris et al., 2020). Attentional biases tend to accompany ACEs/adversity exposure and reflect reduced emotional processing capacity (Davies et al., 2018; 2020a; Nygaard et al., 2016).

Pupillometry

Emotional experiences result in autonomic arousal-related fluctuations suggesting the LC-NE system has an important and direct role in physiological aspects of emotional processing (Olive & Anikin 2018). LC activation reliably predicts changes in pupil diameter on a fine temporal scale both during natural fluctuation and as part of task-driven responding (Joshi et al., 2016). Extensive investigations of pupillary change during affective image viewing suggests pupils increase in diameter when images are both positively and negatively valenced (Bradley et al., 2008)). However, recent work using a task designed to drive attention combined with affective auditory stimuli found pupil dilation occurring faster in the presence of positive and neutral stimuli compared to negative stimuli (Nakakoga et al., 2020). It is important to test for similar effects with emotionally valenced visual stimuli combined with a cognitively effortful task to determine if similar patterns exist naturally in caregivers of children with ACEs and PSE.

Further, it is important to explore the extent that sympathetic activity drives this response compared to parasympathetic contributions to pupil diameter.

Normally it would be difficult to separate autonomic contributions to pupil diameter fluctuations. A study used physiological measures of sympathetic arousal and found underlying increases in sympathetic activity contributing to pupil responses when viewing emotional stimuli regardless of hedonic valence and the use of a task (Bradley et al., 2008). Sympathetic input from LC activation contributes to asymmetry in evoked pupil responses suggesting lateralization in task-evoked changes in pupil diameter may serve as a biomarker of autonomic tone without need for additional measures of autonomic activity (Bradley et al., 2008; Lui et al., 2017).

Cognitive processing also affects pupil size during tasks without emotion-based stimuli suggesting that shifts in pupil diameter when matched with decision criteria provide a means of evaluating the dynamic relationship between emotional/affective processing and arousal state. Additionally, pupil diameter parsed by emotion type provides insight into arousal associated with both emotional fluctuation and cognitive evaluation of emotional stimuli (Oliva & Anikin, 2018). Adversity exposure typically results in increased stress reactivity suggesting that populations with increased ACEs have heightened susceptibility to the effects of future stressors. Differences in LC-NE reactivity between individuals potentially reflects a biomarker of resiliency mechanisms where decreased LC-NE reactivity is protective against chronic stress and ensuing psychopathology (Grueschow et al., 2021).

Purpose and Research Objectives

Eye-tracking provides a novel opportunity to noninvasively measure emotional arousal and indirectly assess LC-NE function. The current study aims to use pupil diameter as a mediator between adversity exposure and negative outcomes to address the role of stress reactivity.

Specifically, the effects of hedonic tone on pupil diameter were measured using eye-tracking as a biomarker of emotional reactivity and processing.

Pupillometry addresses potential differences in the effect of negatively valenced vs. positively valenced stimuli on pupil diameter in caregivers to diversify gaze-point variables and assess autonomic contributions to emotional literacy in caregivers. Increased pupil diameter to negatively valenced stimuli may predict decreased security in the caregiver-child relationship and increased externalizing behaviors in the child. Additionally, we examined whether caregiver pupillary changes act as a mediator for the relationship between child emotional processing strategies and problem behaviors and predict pupillary changes in the child by assessing correlations between biomarkers of emotional attention, emotional literacy, and emotional processing skills defined by the previous evaluation of caregiver and child eye-tracking variables. Beyond aforementioned predictions, analyses were exploratory and therefore hypothesis generating

Methods

Participants

Caregiver-child dyads (N = 30) with children between ages 3 and 7 were recruited from the Child Study Center at the University of Oklahoma Health Sciences Center (OUHSC), which is Oklahoma's only center for trauma intervention and research. Families were patients at A Better Chance Clinic (ABC) and children had a history (or suspected history) of prenatal substance exposure at the time they were recruited to participate. Adult participants are referenced as caregivers regardless of their biological or legal relationship with the child. All caregivers have maintained a relationship with the child for a minimum of 6 months to ensure the

caregiver-child relationship was established with time allowed for caregiving behaviors to affect development.

Materials

A TOBII T120 eye-tracking system measured pupil diameter concurrently with the task indexing emotional processing occurring at the time of intake after clinical testing is complete. For detailed information on stimuli and task set up, see chapter 1.

Procedure

Pupillometry was a secondary analysis from data collected via a Tobii eye-tracking system to assess attention bias as a biomarker for emotional processing capacity. Pupil diameter was recorded at the refresh rate of the Tobii eye-tracking system in millimeters, providing a fine time resolution for dilation speed. Tobii algorithms accounted for the magnification effect from spherical nature of the cornea and the distance to the eye. Written and verbal consent were collected prior to participation in the eye-tracking task from caregivers who provided consent for both caregiver and child participation. Eye-tracking procedural steps are detailed in chapter 1. Eye-tracking recordings took place in a dimly lit room approved for use by the Child Study Center, as the infrared sensors on the Tobii Eye-Tracking system work best in low-lighting and isoluminance conditions are required for use of pupillometry to evaluate effects of NE release from LC (Oliva & Anikin, 2018; Tobii Pro AB, 2012*b*).

Pupillometry

The pupillary data were prepared and analyzed using MATLAB 2020b (Mathworks Inc). Pupil dilation response to target and the 5 non-target images within each trial were averaged within participants separately. Average pupil diameter responses to the 5 non-target images were subtracted from average target pupil diameter responses to compute a general pupillometry

variable (Kinner et al., 2017; Nakakoga et al., 2020). A second pupillometry variable was created for caregivers by generating neutral, anger, fear, and sad pupillometry variables (i.e., subtracting pupil responses to the general trial from emotion-specific targets), subtracting neutral pupil responses from the individual negative image pupil responses, and creating a composite variable to represent pupil diameter responses to negative target images. Pupil diameter was also averaged across all 2500 ms trial periods to create a covariate that controlled for changes in isoluminace as a product of trial brightness (Kinner et al., 2017).

Data analysis

Pupillometry was evaluated in models with attention bias to determine the relationship between LC-NE system function and attention patterns to emotionally valanced stimuli. Pupil diameter, assessed as pupil dilation speed, and changes to emotionally valenced stimuli were compared between eyes to address potential lateralization. We compared intertrial pupil diameter to pupil diameter during trials and then split trials into target vs non target media to assess differences in pupil diameter for all participants (caregivers and children). Correlation analyses addressed relationships between child pupil diameter with the number of externalizing behaviors (ECBI) they exhibited, cognitive capacity, trauma/in-home hostility exposure, and caregiver eye-tracking variables (e.g., emotional processing composite variable, pupillometry, etc.).

Average pupil diameter from each eye for intertrial, target, and nontarget trial images were all normally distributed, but child and caregiver pupillometry variables violated normality assumptions. Child data were normalized by removing obvious outliers (N = 1). Overall caregiver pupillometry and certain caregiver emotion-specific pupillometry variables violated normality. All pupillometry variables were converted to positive values by adding the maximum negative value and 1 then log transforming for interpretability. Caregiver pupil variables were all

log transformed to correct normality violates within certain variables and ensure comparable scale across variables of the same type.

Statistical Analysis

Bootstrapped conditional mediation models were conducted using model 63 the PROCESS macro and examined whether caregiver arousal levels to target via pupillometry and negative media specific pupillometry mediated the relationship between child pupillometry variables and both ECBI and BASC-3 externalizing variables using child age to control for inherent developmental range and trauma exposure as a moderator (Figure 7). Multivariate analyses assessed differences on pupil variables and included intertrial pupil diameter as a covariate to control for pupil changes due solely to screen-based isoluminance changes. Correlations were conducted using Spearman's rho to accommodate violation of normality (i.e., caregiver pupillometry).

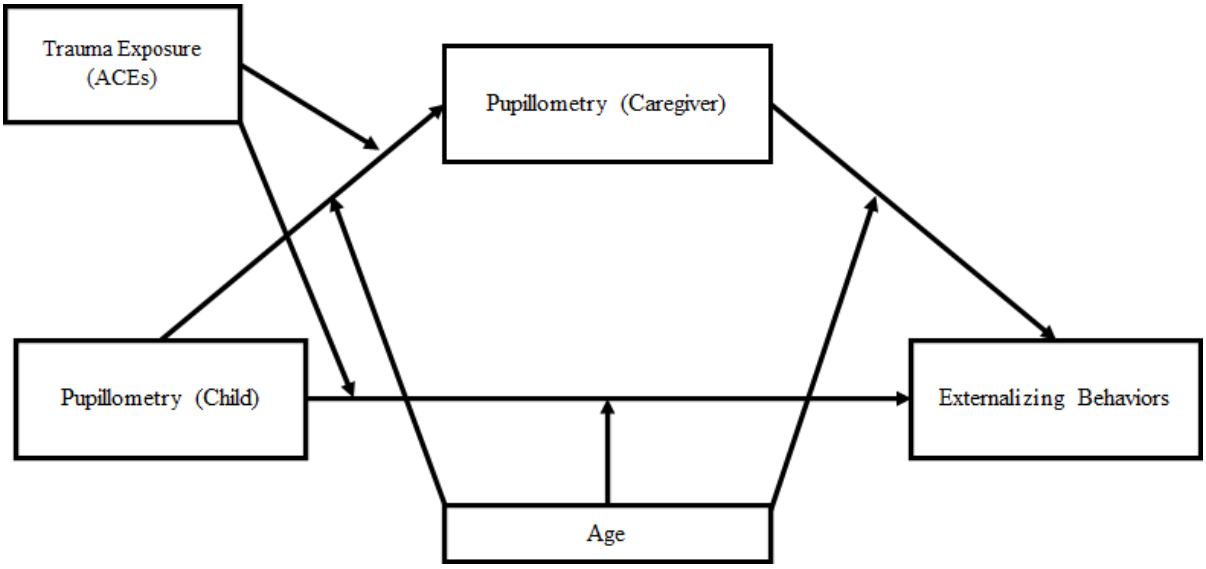

Figure 8. Mediation model for the relationship between child eye-tracking variables and child externalization behaviors. Blank = *N.S.*, *$p < .05$, ** $p < .01$, *** $p < .001$.

Results

Using ECBI intensity scores as the primary dependent variable, the total effect model explained 62.29% of the variability in externalizing behaviors, ($F(7, 9) = 1.51$, $p = .15$, $R^2 = .63$ (Figure 9-10; Table 6a)), with no direct effect of child pupillometry on externalizing behaviors ($\beta = -58.10$, C.I.[-202.96 86.76], $p = .39$) and no indirect effect of caregiver pupillometry on externalizing behaviors ($\beta = -324.78$, C.I.[-862.82 213.26], $p = .21$) (see Appendix D, Figure D1). There was a significant conditional interaction between child pupillometry and age on externalizing behaviors ($\beta = 286.98$, C.I. [10.00 563.95], $p = .044$). The conditional indirect effects of child pupillometry on externalizing behaviors were not significant suggesting no full mediation in the model (Table 6b). There was a significant unconditional interaction between child pupillometry and age ($F(1, 9) = 5.49$, $p = .044$, $\Delta R^2 = .23$), but otherwise no significant unconditional interactions.

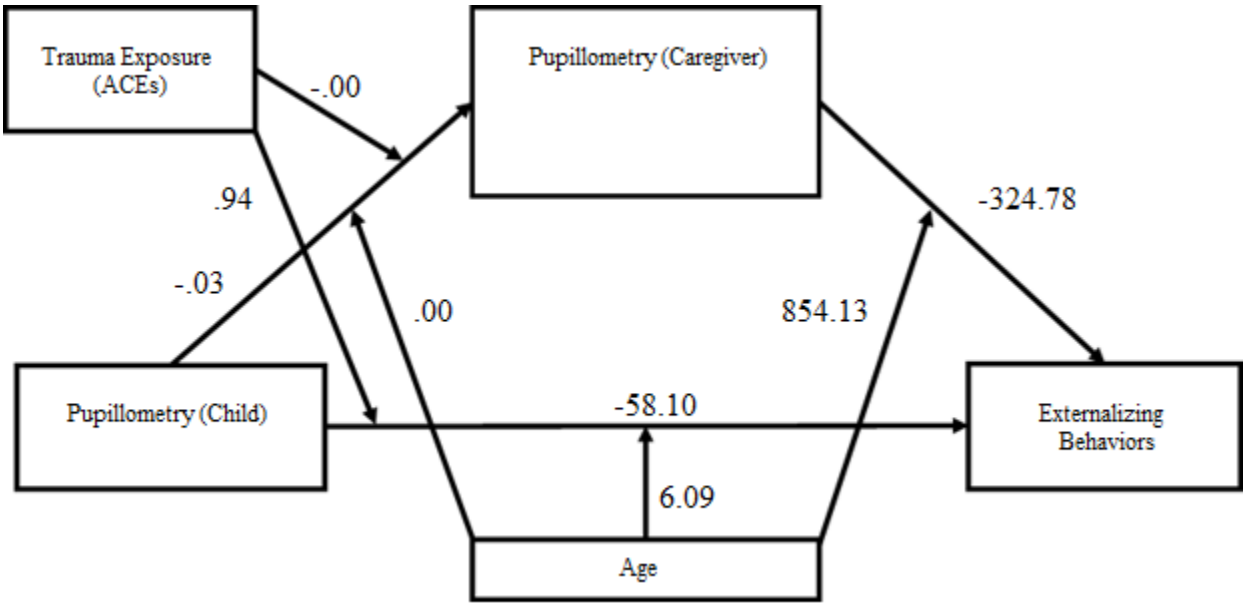

Figure 9. Mediation model and path coefficients for the relationship between child pupillometry and child externalization behaviors measured using ECBI intensity scores. Blank = N.S., *p < .05, ** p < .01, *** p < .001.

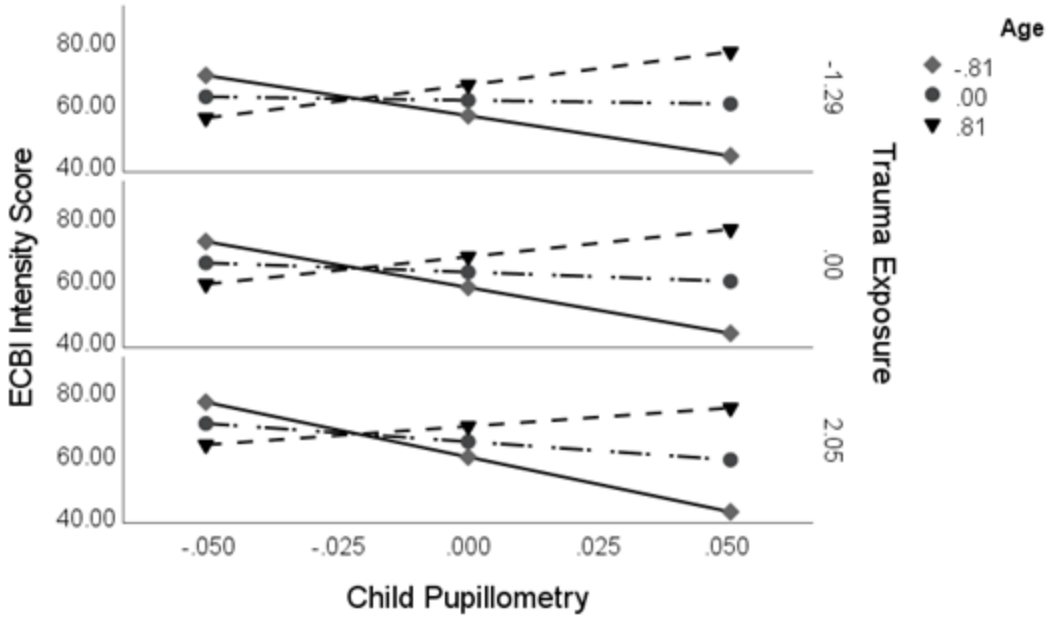

Figure 10. Data visualization for the conditional effect of the focal predictor for pupillometry models with conditioning values at the mean and ±1 standard deviation.

Table 6a. *Conditional Direct Effects of X on Y at levels of the moderators for the Child & General Caregiver Pupillometry Modes using the ECBI Intensity Score*

Age	ACE#	Effect	se	t	p	LLCI	ULCI
-0.81	-1.29	-253.54	100.69	-2.52	0.033	-481.40	-25.68
-0.81	.00	-289.21	116.92	-2.47	0.035	-553.79	-24.62
-0.81	2.05	-345.83	180.09	-1.92	0.087	-753.35	61.70
.00	-1.29	-22.43	72.18	-0.31	0.76	-185.76	140.90
.00	.00	-58.10	64.01	-0.91	0.39	-202.96	86.76
.00	2.05	-114.72	124.45	-0.92	0.38	-396.34	166.89
0.81	-1.29	208.67	140.39	1.49	0.17	-109.03	526.37
0.81	.00	173.00	118.13	1.46	0.18	-94.33	440.33
0.81	2.05	116.38	134.06	0.87	0.41	-187.00	419.76

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Table 6b. *Conditional Indirect Effects of X on Y at levels of the moderators for the Child & General Caregiver Pupillometry Modes using the ECBI Intensity Score*

Age	ACE#	Effect	BootSE	BootLLCI	BootULCI
-0.81	-1.29	123.48	1626.76	-694.16	1091.35
-0.81	0	106.5	165160.2	-901.7	1222.72
-0.81	2.05	79.54	427348.9	-1781.45	1836.45
0	-1.29	14.21	424.86	-145.42	601.83
0	0	8.76	10591.99	-168.24	398.82
0	2.05	0.11	27393.59	-712.94	418.9
0.81	-1.29	12.51	1033.14	-595	918.9
0.81	0	18.6	144161.8	-599.95	750.09
0.81	2.05	28.26	372872.5	-887.22	874.25

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

A second model using BASC-3 externalizing scores for all dyads showed a similar outcome with an overall model that was not significant and explained 37.72% of the variability in externalizing behaviors, $F(7, 21) = 1.82$, $p = .14$, $R^2 = .38$ (Table 9; see Appendix D, Figure D2). There was a nonsignificant direct effect of child pupillometry on externalizing behaviors ($\beta = 15.65$, C.I.[-75.01 106.32], $p = .72$) and the conditional indirect effects of child pupillometry on externalizing behaviors were not significant suggesting no full mediation in the mode (Table 9b).

To assess whether dyads without ECBI scores (i.e., less or no externalizing symptoms) dampened model effects, a third model was conducted on the dyad subset with ECBI intensity scores (excluding the outlier) using BASC-3 externalizing scores as the primary dependent variable. The effect of child pupillometry on externalizing behaviors ($\beta = -13.99$, [-190.42 162.44], $p = .86$) and the overall model were not significant, $F(7, 9) = 1.68$, $p = .23$, $R^2 = .57$ (Table 9). While not significant, the model accounted for 56.60% of the variability in externalizing behaviors (see Appendix D, Figure D3). The conditional indirect effects of child pupillometry on externalizing behaviors were not significant suggesting no full mediation in the model (Table 9b).

A second set of models were tested on a negative target-specific caregiver pupillometry variable. Testing the effect of child pupillometry on externalizing behaviors resulted in non-significant total effect models across all models using ECBI intensity and BASC-3 externalizing scores (Table 7a-b, 9, 9c). Only the ECBI intensity score model had significant effects. The overall model summary was not significant with the model explaining 61.89% of the variability in externalizing behaviors (i.e., ECBI), F (7, 9) = 2.09, p = .15, R^2 = .62 (Figure 11; Table 7a). The conditional indirect effects of child pupillometry on externalizing behaviors were not significant suggesting no full mediation in the model (Table 7b). Age trended toward significance as a conditional moderator of the relationship between child pupillometry and externalizing behaviors (β = 7.33, [-.59 15.26], p = .066), there was a conditional interaction between child pupillometry and age (β = 260.41, [7.95 512.86], p = .044), and the unconditional interaction between child pupillometry and age was significant, F (1, 9) = 5.45, p = .044, ΔR^2 = .23 (see Appendix E).

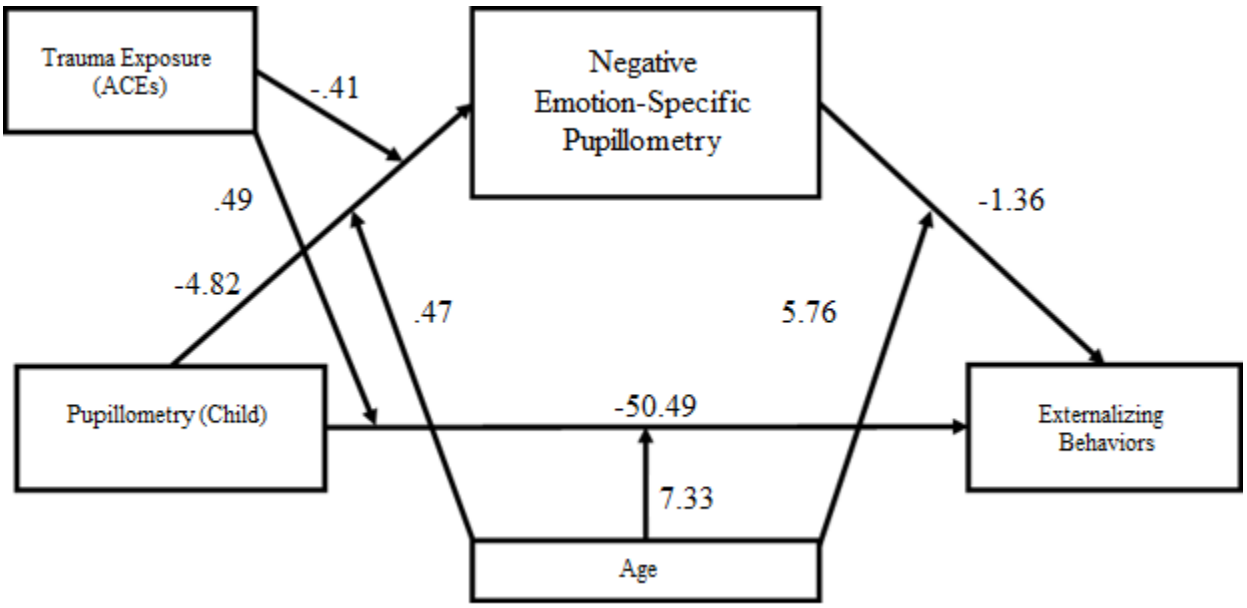

Figure 11. Mediation model and path coefficients for the relationship between child pupillometry and child externalization behaviors measured using ECBI intensity scores with caregiver emotion-specific pupillometry as the mediator. Blank = N.S., *p < .05, ** p < .01, *** p < .001.

Table 7a. *Conditional Direct Effects of X on Y at levels of the moderators for the Negative Emotion-specific Caregiver Pupillometry Model as the Mediator using the ECBI Intensity Score*

Age	ACE#	Effect	se	t	p	LLCI	ULCI
-0.81	-1.29	-234.72	94.87	-2.47	0.04	-449.4	-20.03
-0.81	.00	-260.2	105.57	-2.46	0.04	-499.10	-21.30
-0.81	2.05	-300.66	166.16	-1.81	0.10	-676.68	75.36
.00	-1.29	-25.01	73.49	-0.34	0.74	-191.30	141.28
.00	.00	-50.5	63.90	-0.79	0.45	-195.10	94.10
.00	2.05	-90.95	122.69	-0.74	0.48	-368.59	186.68
0.81	-1.29	184.7	133.95	1.38	0.20	-118.42	487.82
0.81	0	159.21	114.73	1.39	0.20	-100.42	418.84
0.81	2.05	118.75	136.52	0.87	0.41	-190.18	427.68

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Table 7b. *Conditional Indirect Effects of X on Y at levels of the moderators for the Negative Emotion-specific Caregiver Pupillometry Model as the Mediator using the ECBI Intensity Score*

Age	ACE#	Effect	BootSE	BootLLCI	BootULCI
-0.81	-1.29	112.27	2278.08	-778.29	701.25
-0.81	0	95.84	2952.29	-896.03	758.11
-0.81	2.05	69.77	5007.46	-1822.02	1386.96
0	-1.29	10.28	958.45	-264.74	310.16
0	0	6.56	1575.05	-210.51	248.76
0	2.05	0.65	2603.7	-366.05	417.21
0.81	-1.29	11.78	1556.4	-967.7	688.61
0.81	0	20.76	1988.87	-771.31	659.55
0.81	2.05	35.01	2927.12	-844.09	1231.99

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. Boot = bootstrapped, LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

An exploratory conditional mediation model was conducted to assess the potential mediating effect of caregiver composite scores as an index of emotional processing capacity for the relationship between child pupillometry and externalizing behaviors using ECBI intensity scores, motivated by correlations between both caregiver composite scores and ECBI intensity scores with trauma exposure. The model included age and trauma exposure as moderators. With externalizing behaviors (i.e., ECBI intensity scores) as the outcome variable, the total effect model was not significant while still explaining 66.17% of the variability in externalizing behaviors, (F (7, 9) = 2.52, p = .099, R^2 = .66 (Figure 12-13; Table 8a)) with the indirect effect of caregiver composite scores (β = 49.16, [-6.16 216.35], p = .061) and the conditional moderating effect of age on the relationship between child pupillometry and externalizing behaviors (β = 35.09, [-1.24 71.43], p = .057) trending toward significance (see Appendix F, Figure F1). The conditional indirect effects of child pupillometry on externalizing behaviors were not significant suggesting no full mediation in the model (Table 8b). The simple regression model assessing the relationship between caregiver composite scores and child externalizing behaviors showed a trending outcome with the model explaining 55.83% of the variability in

caregiver composite scores, F (5, 11) = 2.78, p = .073, R^2 = .56. The relationship between child pupillometry and caregiver composite scores was significant (β = -3.99, [-7.57 -.40], p = .032), but no other direct effects or interactions were significant.

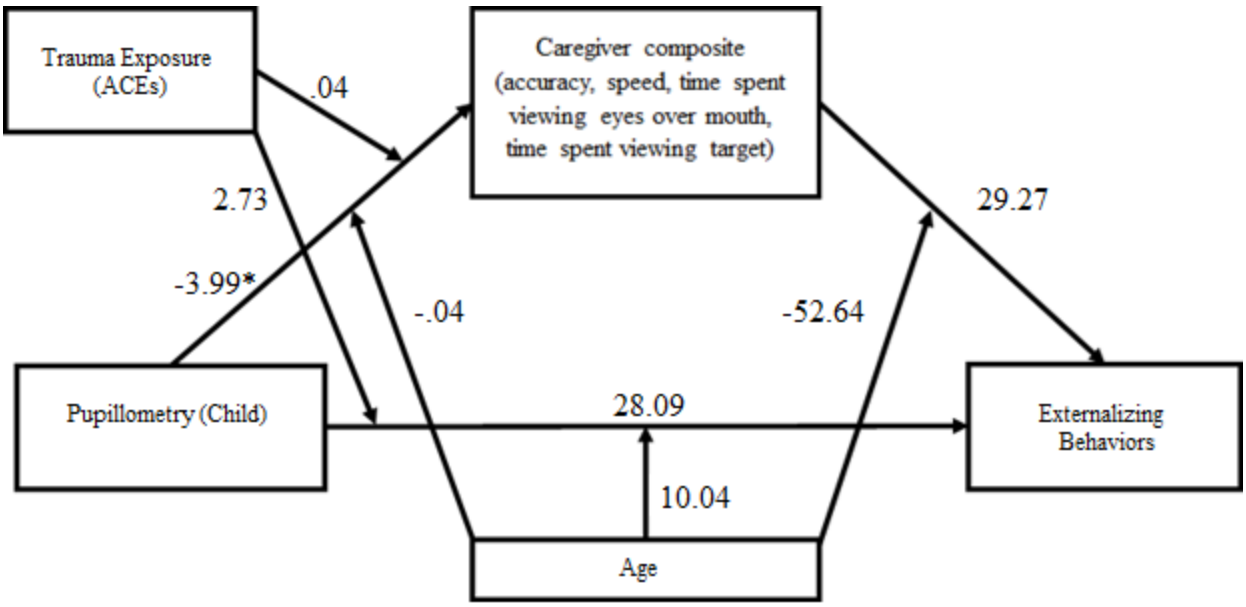

Figure 12. Mediation model and path coefficients for the relationship between child pupillometry and child externalization behaviors measured using ECBI intensity scores with caregiver composite scores as the mediator. Blank = N.S., *p < .05, ** p < .01, *** p < .001.

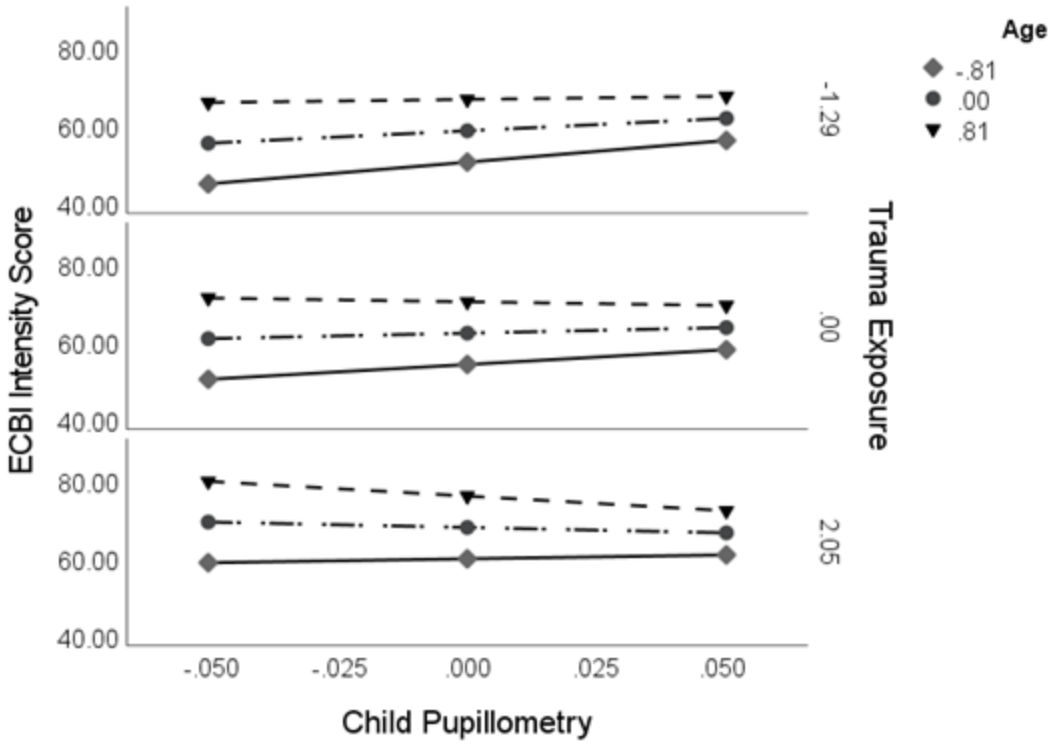

Figure 13. Data visualization for the conditional effect of the focal predictor for the pupillometry model with caregiver composite scores as the mediator. Conditioning values reflect the mean and ±1 standard deviation.

Table 8a. *Conditional Direct Effects of X on Y at levels of the moderators for the Pupillometry Model with Caregiver Composite Scores as the Mediator using the ECBI Intensity Score*

Age	ACE#	Effect	se	t	p	LLCI	ULCI
-0.81	-1.29	110.28	123.07	0.9	0.39	-168.21	388.78
-0.81	0	75.31	119.06	0.63	0.54	-194.1	344.73
-0.81	2.05	19.79	164.93	0.12	0.91	-353.43	393.01
0	-1.29	63.06	76.22	0.83	0.43	-109.42	235.55
0	0	28.09	74.45	0.38	0.71	-140.39	196.58
0	2.05	-27.43	140.32	-0.2	0.85	-344.96	290.11
0.81	-1.29	15.84	144.01	0.11	0.91	-310.05	341.74
0.81	0	-19.13	145.53	-0.13	0.90	-348.45	310.19
0.81	2.05	-74.65	190.9	-0.39	0.70	-506.65	357.35

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Table 8b. *Conditional Indirect Effects of X on Y at levels of the moderators for the Pupillometry Model with Caregiver Composite Scores as the Mediator using the ECBI Intensity Score*

Age	ACE#	Effect	BootSE	BootLLCI	BootULCI
-0.81	-1.29	-246.34	3672.5	-1681	595.31
-0.81	0	-333.3	119843.4	-2638.64	767.67
-0.81	2.05	-471.35	310033.9	-4710.7	1478.31
0	-1.29	-80.98	538.71	-473.68	120.29
0	0	-116.68	35705.98	-768.8	202.87
0	2.05	-173.35	92368.03	-1616.52	526.34
0.81	-1.29	26.52	1218.75	-787.38	606.14
0.81	0	42.08	48494.31	-727.58	773.26
0.81	2.05	66.79	125384.1	-843.67	1347.42

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. Boot = bootstrapped, LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Assessing the same model using BASC-3 externalizing scores as the externalizing behaviors measure for all dyads resulted in a model outcome that was not significant and explained only 38.74% of the variability in externalizing behaviors with only a trending moderating effect of ACEs on the relationship between child pupillometry and caregiver composite scores (β = -.05, [-.11 .003], p = .064), F (7, 21) = 1.89, p = .12, R^2 = .39 (Table 9; see Appendix F, Figure F2). The conditional indirect effects of child pupillometry on externalizing behaviors were not significant suggesting no mediation in the model (Table 9d).

Assessing the BASC-3 externalizing model with the dyad subset resulted in the total effect trending toward significance with the model explaining 70.12% of the variability in externalizing behaviors, F (7, 9) = 3.02, p = .063, R^2 = .70 (Table 9; see Appendix F, Figure F3). The conditional indirect effects of child pupillometry on externalizing behaviors were not significant suggesting no mediation in the model (Table 9d). The relationship between caregiver composite scores and externalizing behaviors (β = 33.24, [-.19 66.67], p = .051), the moderating effect of age on the relationship between child pupillometry and externalizing behaviors (β = 9.97, [-.94 20.89], p = .069), and The simple regression model assessing relationships with

caregiver composite scores trending toward significance and explained 55.83% of the variability in caregiver composite scores, F (5, 11) = 2.78, p = .073, R^2 = .56, and a significant direct effect of child pupillometry on caregiver composite scores was present (β = -3.99, [-7.57 -.40], p = .032). No unconditional interactions were significant.

Table 9. *Pupillometry Mediation Model Path Coefficients for Models using BASC-3 Externalizing Scores as the Externalizing Behavior Variable.*

Model Path	Standardized path coefficients	
	BASC-3 Ext All dyads	**BASC-3 Ext Dyad subset (N = 17)**
Caregiver General Pupillometry		
CPPL → CGPPL	-.06	-.03
Age: CPPL → CGPPL	.003	.001
ACEs: CPPL → CGPPL	-.0003	-.002
CPPL x Age→ CGPPL	-.03	.09
CPPL x ACEs → CGPPL	-.002	.01
CGPPL → Externalizing behaviors	-.85	-233.99
CPPL → Externalizing behaviors	15.65	-13.99
Age: CPPL → Externalizing behaviors	3.95	5.07
ACEs: CPPL → Externalizing behaviors	-1.36	2.66
CPPL x Age → Externalizing behaviors	12.75	271.03
CGPPL x Age → Externalizing behaviors	-401.97	288.45
CPPL x ACEs → Externalizing behaviors	41.70	-80.87
Caregiver Emotion-Specific Pupillometry		
CPPL → CGPPL	-2.06	-4.82
Age: CPPL → CG ESPPL	.19	.47
ACEs: CPPL → CG ESPPL	-.22	-.41
CPPL x Age→ CG ESPPL	1.09	13.84
CPPL x ACEs → CG ESPPL	.55	2.11
CG ESPPL → Externalizing behaviors	-.50	-.56
CPPL → Externalizing behaviors	26.23	-10.71
Age: CPPL → Externalizing behaviors	3.64	5.72
ACEs: CPPL → Externalizing behaviors	-1.11	2.67
CPPL x Age → Externalizing behaviors	45.97	267.32
CCS x Age → Externalizing behaviors	.02	2.68
CPPL x ACEs → Externalizing behaviors	38.56	-80.03
Caregiver Composite Score		
CPPL → CCS	-.97	-3.99*
Age → CCS	-.002	-.04
ACEs → CCS	-.05	.04
CPPL x Age→ CCS	-1.59	.85
CPPL x ACEs → CCS	.74	-.94
CCS → Externalizing behaviors	18.53	33.24
CPPL → Externalizing behaviors	44.17	73.48
Age: CPPL → Externalizing behaviors	3.64	9.97
ACEs: CPPL → Externalizing behaviors	-.019	3.96
CPPL x Age → Externalizing behaviors	74.13	28.24
CCS x Age → Externalizing behaviors	-.95	-50.04
CPPL x ACEs → Externalizing behaviors	24.21	-82.29

Note. $*p < 0.05$, $**p < 0.01$, $***p < 0.001$. Mediation models all follow the same model structure as those in **Figure 7**. Listed model paths are organized by the mediating variable in each model. Age and ACEs are moderators. Abbreviations: ACEs = trauma exposure, CCS = caregiver composite score, CG = caregiver, CPPL = child pupillometry, ES = emotion-specific, PPL = pupillometry.

Table 9b. *Conditional Indirect Effects of X on Y at levels of the moderators for Pupillometry with General Caregiver Pupillometry as Mediator using the BASC-3 Externalizing Score*

General Caregiver Pupillometry

Age	ACE#	Effect	Boot SE	Boot LLCI	Boot ULCI
BASC-3 Externalizing All					
-0.93	-1.59	-5.72	62.65	-182.62	63.17
-0.93	0	-6.38	49.89	-151.04	50.15
-0.93	1.82	-7.13	53.96	-144.48	67.25
0	-1.59	7.92	41.95	-38.9	119.74
0	0	8.31	28.61	-22.08	86.91
0	1.82	8.76	25.52	-24.3	74.91
0.93	-1.59	45.54	132.24	-68.5	449.93
0.93	0	46.98	111.59	-51.43	375.23
0.93	1.82	48.63	105.26	-60.25	351.85
BASC-3 Externalizing Subset					
-0.81	-1.29	56.86	1006.2	-755.48	1015.56
-0.81	0	49.04	1467.9	-1098.26	1269.89
-0.81	2.05	36.63	2958.83	-1917.75	2240.31
0	-1.29	10.23	357.07	-181.89	691.35
0	0	6.31	322.6	-186.28	464.25
0	2.05	0.08	659.44	-762.56	560.46
0.81	-1.29	-0.06	1475.79	-776.54	1171.48
0.81	0	-0.09	1177.99	-717.45	890.57
0.81	2.05	-0.13	1653.57	-1210.73	965.57
-0.81	-1.29	56.86	1006.2	-755.48	1015.56

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Table 9c. *Conditional Indirect Effects of X on Y at levels of the moderators for Pupillometry with Negative Emotion-Specific Caregiver Pupillometry as Mediator using the BASC-3 Externalizing Score*

Negative Emotion-Specific Caregiver Pupillometry

Age	ACE#	Effect	Boot SE	Boot LLCI	Boot ULCI
BASC-3 Externalizing All					
-0.93	-1.59	2.06	65.64	-112.28	165.19
-0.93	0	1.61	48.91	-72.88	124.92
-0.93	1.82	1.08	57.78	-81.96	138.85
0	-1.59	1.47	24.37	-38.72	63.87
0	0	1.03	18.09	-26.47	44.61
0	1.82	0.53	23.13	-37.44	59.32
0.93	-1.59	0.92	67.42	-149.21	133.74
0.93	0	0.5	63.92	-140.14	121.47
0.93	1.82	0.02	71.29	-166.11	131.96
BASC-3 Externalizing Subset					
-0.81	-1.29	51.02	1773.96	-1088.87	764.11
-0.81	0	43.56	4914.21	-1021.3	1009.66
-0.81	2.05	31.71	14164.39	-1768.92	1968.91
0	-1.29	4.27	2308.83	-350.97	333.69
0	0	2.72	1990.25	-271.05	302.61
0	2.05	0.27	6086.98	-504.6	648.06
0.81	-1.29	5.74	5268.06	-1004.36	918.88
0.81	0	10.11	3996.78	-864.41	772.9
0.81	2.05	17.05	3719.3	-1125.34	1286.72
-0.81	-1.29	51.02	1773.96	-1088.87	764.11

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Table 9d. *Conditional Indirect Effects of X on Y at levels of the moderators for Pupillometry with Caregiver Composite Scores as Mediator using the BASC-3 Externalizing Score*

Pupillometry with Caregiver Composites

Age	ACE#	Effect	Boot SE	Boot LLCI	Boot ULCI
BASC-3 Externalizing All					
-0.93	-1.59	-12.71	41.25	-113.82	58.33
-0.93	0	10.03	31.31	-59.13	71.48
-0.93	1.82	36.16	53.17	-62.99	149.74
0	-1.59	-39.63	41.03	-141.3	21.16
0	0	-17.92	25.71	-82.76	22.25
0	1.82	7.03	35.38	-71.4	79.17
0.93	-1.59	-63.96	109.74	-338.67	81.71
0.93	0	-43.27	88.65	-277.82	76.69
0.93	1.82	-19.5	84.24	-259.72	100.96
BASC-3 Externalizing Subset					
-0.81	-1.29	-254.1	1666.39	-1628.45	753.25
-0.81	0	-343.81	5059.68	-2866.57	960.98
-0.81	2.05	-486.21	11460.43	-4864.3	1432.67
0	-1.29	-91.96	295.56	-437.42	144.17
0	0	-132.51	1448.45	-801.62	205.15
0	2.05	-196.87	3526.37	-1601.15	489.14
0.81	-1.29	14.69	816.75	-924.29	481.02
0.81	0	23.31	1661.63	-854.37	692.48
0.81	2.05	36.99	4226.01	-1042.11	1179.84
-0.81	-1.29	-254.1	1666.39	-1628.45	753.25

Note. Moderators were split on conditioning values at the mean and +/- 1 standard deviation at a 95% CI. LLCI = lower limit confidence interval, ULCI = upper limit confidence interval.

Pupillometry Clinical Correlations

Correlations of note include direct relationships between caregiver pupillometry and both child pupil diameter to targets (rho = .31, *p* = .003) and to trials (rho = .35, *p* = .002), but not with child pupillometry, rho = -.25, *p* = .19. Neither caregiver nor child pupillometry were correlated with caregiver composite scores (Figure 14). For the dyads included in pupillometry analyses, there was a correlation between trauma exposure and caregiver composites scores, rho = -.51, *p* = .003 (Figure 10).

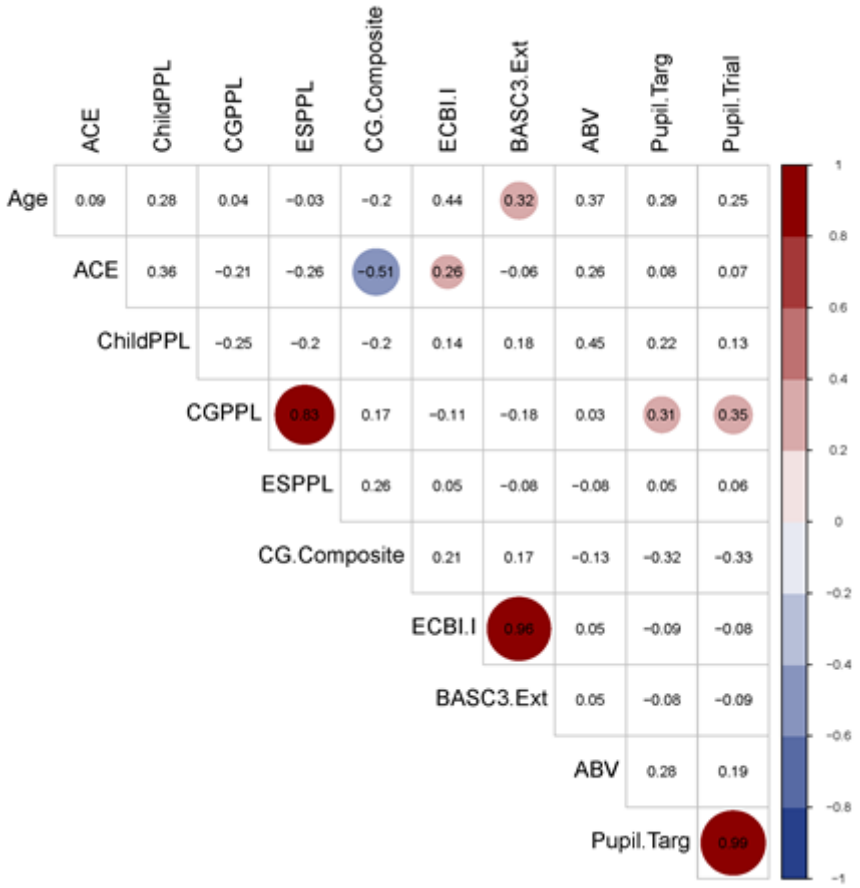

Figure 14. All correlations are Spearman's rho. All significant correlations are listed on colored circles. Blank = N.S. Abbreviations: ABV = general attention bias, ACE = trauma exposure, CG = caregiver, PPL = pupillometry, Targ = target image viewing, Ext = externalizing behavior, I = intensity.

Child-Specific Pupil Results

A two-way repeated measure ANOVA was conducted to assess condition by eye differences in pupil dilation. Specifically, we tested for differences in pupil dilation when attending to the target compared to other faces within the trial (i.e., condition) with intertrial pupil diameter included as a covariate to control for isoluminance. The main effect of eye (F(1, 27) = .012, p = .92, ES = .00) and condition (F(1, 27) = .94, p = .76, ES = .95) were not significant, but intertrial pupil diameter was a significant covariate suggesting significant pupil constriction occurred as an effect of trial onset, F(1, 27) = 539.8, p < .001, ES = .95.

Caregiver-Specific Pupil Results

A one-way repeated measure ANOVA was conducted to assess overall pupil diameter during target viewing compared to pupil diameter when viewing the nontarget images within a trial (i.e., condition) with intertrial as a covariate to control for changes in isoluminance associated with the onset of the trial. The main effect of condition was not significant (F(1, 28) = .23, p = .64, ES = .01), but intertrial was a significant covariate, F(1, 28) = 498.46, p > .001, ES = .95. A second one-way repeated measure ANOVA was conducted to assess differences across emotion-specific pupillometry variables (i.e., anger-, sad-, fear-, and neutral-specific pupillometry). A significant main effect of emotion was found, F(3, 27) = 5.68, p = .004, ES = .39. Differences were found between fear (M = .03, SD = .00) and both sad (M =.04, SE = .01) and neutral-specific pupillometry (M = .04, SE = .00) with fear-specific pupillometry significantly reduced in comparison.

Discussion

Pupillometry as a measure of LC-NE reactivity within populations with PSE and ACEs reflects a novel approach to evaluating factors that influence increased externalizing behaviors. Current results demonstrate pupillometry measuring arousal fluctuations specific to negatively valenced stimuli as it relates to externalizing behaviors only though an index of caregiver emotional processing. Partial mediation of the relationship between LC-NE reactivity and externalizing behaviors by caregiver emotional processing predicts decreased security in the caregiver-child relationship and provides a measure of environmental biobehavioral feedback.

LC-NE Function Mediation Models

All models demonstrated model relationships where arousal prompted by target viewing did not explain a significant amount of variability in externalizing behaviors, even for children with externalizing behavior problems notable enough for caregivers to report to a clinical team. Further, child pupillometry as an index of LC-NE function as a predictor of externalizing behaviors was not moderated by trauma exposure suggesting a more complex relationship between child adversity exposure and behavior. Model outcomes were different when assessing caregiver composite scores over caregiver pupillometry, such that the relationship between child pupillometry and caregiver composite scores (i.e., caregiver emotional processing capacity) was significant when both ECBI intensity scores and BASC-3 externalizing scores from the dyad subsets were the outcome variable. The relationship between child pupillometry and caregiver composite scores suggests biobehavioral factors related to underlying LC-NE system function in children impact caregivers emotional processing capacity, specifically in dyads where the child exhibited significant externalizing behavior problems (Hofer, 1994). Results for full mediation within all models were not significant despite significant path outcomes suggesting mediation of

the relationship between child pupillometry and externalizing behaviors. Mediation results likely reflect evaluation of a complex statistical model with a small sample size. Current results suggest caregiver emotional processing explains the relationship between child LC-NE reactivity and externalizing behaviors with increases in child pupillometry associated with decreases in caregiver emotional processing, despite a lack of significance for full mediation. With underlying changes in LC-NE function, children likely exhibit behavioral products of LC-NE dysregulation potentially driving the way caregivers process emotions (Hofer, 1994; Saxbe et al., 2016). Caregivers are known to influence the developmental outcomes of their children, with caregiver-child interactions providing scaffolding for important developmental functions. Current results demonstrated increased arousal with decreased caregiver emotional processing capacity indexed via composite score. The direction of this relationship is unknown with a potential for bidirectional biobehavioral impact. Bidirectionality would suggest the child influences caregiver emotional processing capacity and vice versa suggesting the potential for bidirectional biobehavioral impact acting as an environmental feedback loop between caregiver and child (Meyer et al., 2021). Biobehavioral synchrony posits a mechanism where dynamic interactions between caregivers and children that act upon physiological dysregulation modulating emotional and physiological fluctuations (Feldman et al., 2007). Even if caregiver-child relationships are not maladapted, reduced emotional processing capacity in the caregiver feeds into a child's ability to calibrate threat responding as a product of flexibility, susceptibility, and resilience. Dysregulation of threat processing pathways leads to reactive coping which involves elevated HPA-axis activity and decreased emotional regulation (Meyer et al., 2021).

Early life stress and adversity exposure have pervasive effects on frontal and limbic structures, specifically pathways spanning prefrontal-hypothalamic-amygdala areas and

dopaminergic circuits within those pathways. Changes to frontal and limbic structures are partially mediated by changes to HPA-axis function and the autonomic nervous system (Smith & Pollak, 2020). Chronic exposure to adversity or prolonged periods of adversity are known to lead to increases in LC-NE reactivity (Anda et al., 2006). Changes to LC-NE reactivity impacts higher executive functions, including those associated with connections between LC and limbic structures where ACEs/adversity exposure negatively impact performance requiring higher executive functions (Davies et al., 2018; 2020a; Morris et al., 2020; Nygaard et al., 2016). Dysregulation of autonomic nervous systems and atypical HPA-axis function were expected to relate directly to behavior, supported by evidence suggesting HPA-axis dysregulation alters perception of threat and challenges presented by the environment, but trauma exposure did not significantly moderate the relationship between pupillometry and externalizing behavior (Smith & Pollak, 2020). Quantification of adversity exposure for the current study may partially explain these results because chronic and intermittent adversity exposure exhibit different developmental effects (Evans et al., 2013; Schroeder et al., 2021).

Pupillometry as a biomarker potentially indexes both stress pathway reactivity and resiliency. While increased LC-NE reactivity tends to predict negative outcomes, variability in LC-NE function can also predict potential resiliency with decreased reactivity acting as a buffer between adversity experiences and subsequent chronic stress/stress reactivity and psychopathology (Grueschow et al., 2021). ACEs and trauma are broadly investigated with known negative developmental outcomes; however, limited work has aimed to address the physiological mechanisms underlying disruptions in emotional or social processes to establish biomarkers of ACEs. In a clinical population of trauma exposed individuals, there is an increased likelihood that modifications to the stress pathway have occurred resulting in notable arousal

responses to mild or harmless stimuli (e.g., increased HPA axis or LC-NE reactivity). Modifications of neural pathways resulting in atypical reactivity of components in the stress pathway reflect a unique opportunity to address the effects of trauma and the potential role of caregivers on child LC-NE function using tasks to probe specific responses to mild stressors (Grueschow et al., 2021).

Correlation outcomes assessed relationships between clinical measures and pupil diameter as a function of LC-NE reactivity in response to valanced stimuli. The lack of correlation between both caregiver pupillometry variable and the composite scores suggests that pupillometry does not index task performance in the same way the composite score indexes task performance. However, general caregiver pupillometry likely indexes arousal to task performance rather than arousal to specific valenced stimuli. General pupillometry included neutral target identification, likely dampening the ability to measure pupil diameter changes in response to valenced stimuli. The emotion-specific variable still measured target viewing compared to all other images within a trial potentially impacting effect. We predicted direct relationships between increased pupil diameter in response to negatively valanced stimuli with trauma exposure and an inverse relationship between increased child pupil diameter to negatively valanced stimuli with variables indexing caregiver emotional literacy. These relationships were not present, but mediation outcomes suggest these factors interact in a nuanced way that influences child behavior. Further, correlations between caregiver pupillometry and child pupil diameter response to both targets and trial images suggests increased caregiver arousal shares a relationship with increased emotional arousal in the child.

Conclusions

Caregiver emotional processing, but not arousal, may mediate the relationship between child arousal responses to negatively valenced emotional stimuli and externalizing behaviors. Findings suggest shifts in child LC-NE reactivity dynamically interact within the caregiver-child relationship to produce behavioral effects. In conclusion, caregivers are an important developmental and environmental context for child development in populations that have PSE and other early life stressors. As a result, child pupillometry may serve as a biomarker of LC-NE dysregulation with predictive power for externalizing behavior problems.

Limitations and Assumptions

Pupillometry is an indirect measure of LC-NE function and therefore an indirect measure of stress reactivity (Grueschow et al., 2021; Oliva & Anikin, 2018). The LC-NE drives arousal and affective viewing is known to induce arousal responses in populations without adversity exposure, posing a primary limitation on using pupillometry in commentary on the relationship between experiences of early adversity, consequences of adversity exposure, and anatomical/physiological modifications to the arousal pathway.

Affective image viewing is limited to faces making emotional expression ranging from positive to negative. Caregivers were asked to identify the emotion corresponding to a word indicating one of six emotions making the task cognitively effortful but does not provide any conflict related stress as a typical emotional Stroop task would provide (Grueschow et al., 2021). Caregiver pupillometry may better index child-specific responses in arousal if stimuli included child faces. Further, pupillometry calculations may benefit from within trial comparisons between negative and neutral stimuli that were not included in the current study due to concerns about data redundancy effects and skewed standard deviations.

It is well known that ACEs and adversity exposure are linked to alterations in the LC-NE system and recent work has increasingly employed pupillometry to index LC-NE activity. However, the use of pupillometry to evaluate LC-NE function in a population with ACEs and PSE remains novel. The novelty of the current approach is an advantage, but pupil diameter changes in the presence of affective stimuli as a function of LC-NE reactivity should be interpreted with great care.

References

Anda, R. F., Felitti, V. J., Bremner, J. D., Walker, J. D., Whitfield, C., Perry, B. D., Dube, S. R., & Giles, W. H. (2006). The enduring effects of abuse and related adverse experiences in childhood. A convergence of evidence from neurobiology and epidemiology. *Eur Arch Psychiatry Clin Neurosci*, *256*(3), 174-186. https://doi.org/10.1007/s00406-005-0624-4

Atzaba-Poria, N., Deater-Deckard, K., & Bell, M. A. (2017). Mother-Child Interaction: Links Between Mother and Child Frontal Electroencephalograph Asymmetry and Negative Behavior. *Child Dev*, *88*(2), 544-554. https://doi.org/10.1111/cdev.12583

Ballarotto, G. (2016). *Parent-Child Interactions in Families with Mothers who had Experienced Early Relational Traumas*

Bethell, C. D., Carle, A., Hudziak, J., Gombojav, N., Powers, K., Wade, R., & Braveman, P. (2017). Methods to Assess Adverse Childhood Experiences of Children and Families: Toward Approaches to Promote Child Well-being in Policy and Practice. *Acad Pediatr*, *17*(7S), S51-S69. https://doi.org/10.1016/j.acap.2017.04.161

Bick, J., & Nelson, C. A. (2016). Early Adverse Experiences and the Developing Brain. *Neuropsychopharmacology*, *41*(1), 177-196. https://doi.org/10.1038/npp.2015.252

Boggs, S. R., Eyberg, S., & Reynolds, L. A. (2010). Concurrent Validity of the Eyberg Child Behavior Inventory. *Journal of Clinical Child Psychology*, *19*(1), 75-78. https://doi.org/10.1207/s15374424jccp1901_9

Bozkurt Yükçü, Ş., & Demircioğlu, H. (2020). Examining the predictor effect of parents' emotional literacy level on the emotion regulation and social problem-solving skills of children. *Early Child Development and Care*, 1-16. https://doi.org/10.1080/03004430.2020.1720671

Bradley, M. M., Miccoli, L., Escrig, M. A., & Lang, P. J. (2008). The pupil as a measure of emotional arousal and autonomic activation. *Psychophysiology*, *45*(4), 602-607. https://doi.org/10.1111/j.1469-8986.2008.00654.x

Briggs-Gowan, M. J., Pollak, S. D., Grasso, D., Voss, J., Mian, N. D., Zobel, E., McCarthy, K. J., Wakschlag, L. S., & Pine, D. S. (2015). Attention bias and anxiety in young children exposed to family violence. *J Child Psychol Psychiatry*, *56*(11), 1194-1201. https://doi.org/10.1111/jcpp.12397

Buehler, C., Anthony, C., Krishnakumar, A., Stone, G., Gerard, J., & Pemberton, S. (1997). Interparental Conflict And Youth Problem Behaviors. *Journal of Child and Family Studies*, *6*(2), 233-247. https://doi.org/10.1023/A:1025006909538

Cervin, M., Salloum, A., Ruth, L. J., & Storch, E. A. (2021). Posttraumatic Symptoms in 3-7 Year Old Trauma-Exposed Children: Links to Impairment, Other Mental Health Symptoms, Caregiver PTSD, and Caregiver Stress. *Child Psychiatry Hum Dev*, *52*(6), 1173-1183. https://doi.org/10.1007/s10578-020-01093-3

Crespo, L. M., Trentacosta, C. J., Aikins, D., & Wargo-Aikins, J. (2017). Maternal Emotion Regulation and Children's Behavior Problems: The Mediating Role of Child Emotion Regulation. *Journal of Child and Family Studies*, *26*(10), 2797-2809. https://doi.org/10.1007/s10826-017-0791-8

Davies, P. T., Coe, J. L., Hentges, R. F., Sturge-Apple, M. L., & Ripple, M. T. (2018). Interparental hostility and children's externalizing symptoms: Attention to anger as a mediator. *Dev Psychol*, *54*(7), 1290-1303. https://doi.org/10.1037/dev0000520

Davies, P. T., Thompson, M. J., Hentges, R. F., Coe, J. L., & Sturge-Apple, M. L. (2020). Children's attentional biases to emotions as sources of variability in their vulnerability to

interparental conflict. *Dev Psychol*, *56*(7), 1343-1359. https://doi.org/10.1037/dev0000994

Davies, P. T., Thompson, M. J., Martin, M. J., & Cummings, E. M. (2021). The Vestiges of Childhood Interparental Conflict: Adolescent Sensitization to Recent Interparental Conflict. *Child Dev*, *92*(3), 1154-1172. https://doi.org/10.1111/cdev.13451

De Bellis, M. D., Baum, A. S., Birmaher, B., & Ryan, N. D. (1997). Urinary catecholamine excretion in childhood overanxious and posttraumatic stress disorders. *Ann N Y Acad Sci*, *821*, 451-455. https://doi.org/10.1111/j.1749-6632.1997.tb48303.x

Eiden, R. D., Godleski, S., Schuetze, P., & Colder, C. R. (2015). Prenatal substance exposure and child self-regulation: Pathways to risk and protection. *J Exp Child Psychol*, *137*, 12-29. https://doi.org/10.1016/j.jecp.2015.02.007

Erriu, M. (2017). *Longitudinal Study On Parental Psychopathological Risk And Offspring's Growth Delay*

Evans, G. W., Li, D., & Whipple, S. S. (2013). Cumulative risk and child development. *Psychol Bull*, *139*(6), 1342-1396. https://doi.org/10.1037/a0031808

Feldman, R. (2007). Parent infant synchrony: Biological foundations and developmental outcomes. *Current Directions in Psychological Science*, *16*(6), 340-345.

Felitti, V. J., Anda, R. F., Nordenberg, D., Williamson, D. F., Spitz, A. M., Edwards, V., Koss, M. P., & Marks, J. S. (1998). Relationship of childhood abuse and household dysfunction to many of the leading causes of death in adults. The Adverse Childhood Experiences (ACE) Study. *Am J Prev Med*, *14*(4), 245-258. https://doi.org/10.1016/s0749-3797(98)00017-8

Fox, S. E., Levitt, P., & Nelson, C. A., 3rd. (2010). How the timing and quality of early experiences influence the development of brain architecture. *Child Dev*, *81*(1), 28-40. https://doi.org/10.1111/j.1467-8624.2009.01380.x

Fritz, J., de Graaff, A. M., Caisley, H., van Harmelen, A. L., & Wilkinson, P. O. (2018). A Systematic Review of Amenable Resilience Factors That Moderate and/or Mediate the Relationship Between Childhood Adversity and Mental Health in Young People. *Front Psychiatry*, *9*, 230. https://doi.org/10.3389/fpsyt.2018.00230

Goldsmith, D. F., Oppenheim, D., & Wanlass, J. (2004). Separation and reunification: Using Attachment Theory and Research to Inform Decisions Affecting the Placements of Children in Foster Care. *Juvenile and Family Court Journal*, *55*(2), 1-13. https://doi.org/10.1111/j.1755-6988.2004.tb00156.x

Griffin, J. W., & Scherf, K. S. (2020). Does decreased visual attention to faces underlie difficulties interpreting eye gaze cues in autism? *Mol Autism*, *11*(1), 60. https://doi.org/10.1186/s13229-020-00361-2

Grueschow, M., Stenz, N., Thorn, H., Ehlert, U., Breckwoldt, J., Brodmann Maeder, M., Exadaktylos, A. K., Bingisser, R., Ruff, C. C., & Kleim, B. (2021). Real-world stress resilience is associated with the responsivity of the locus coeruleus. *Nat Commun*, *12*(1), 2275. https://doi.org/10.1038/s41467-021-22509-1

Hayes, A. F. (2017). Hacking process for estimation and probing of linear moderation of quadratic effects.

Herzog, J. I., & Schmahl, C. (2018). Adverse Childhood Experiences and the Consequences on Neurobiological, Psychosocial, and Somatic Conditions Across the Lifespan. *Front Psychiatry*, *9*, 420. https://doi.org/10.3389/fpsyt.2018.00420

Hofer, M. A. (1994). Hidden regulators in attachment, separation, and loss. *Monographs of the Society for Research in Child Development*, *59*(2/3), 192-207.

Joshi, S., Li, Y., Kalwani, R. M., & Gold, J. I. (2016). Relationships between Pupil Diameter and Neuronal Activity in the Locus Coeruleus, Colliculi, and Cingulate Cortex. *Neuron*, *89*(1), 221-234. https://doi.org/10.1016/j.neuron.2015.11.028

Kauschke, C., Bahn, D., Vesker, M., & Schwarzer, G. (2019). The Role of Emotional Valence for the Processing of Facial and Verbal Stimuli-Positivity or Negativity Bias? *Front Psychol*, *10*, 1654. https://doi.org/10.3389/fpsyg.2019.01654

Kinner, V. L., Kuchinke, L., Dierolf, A. M., Merz, C. J., Otto, T., & Wolf, O. T. (2017). What our eyes tell us about feelings: Tracking pupillary responses during emotion regulation processes. *Psychophysiology*, *54*(4), 508-518. https://doi.org/10.1111/psyp.12816

Knopik, V. S., Marceau, K., Bidwell, L. C., & Rolan, E. (2019). Prenatal substance exposure and offspring development: Does DNA methylation play a role? *Neurotoxicol Teratol*, *71*, 50-63. https://doi.org/10.1016/j.ntt.2018.01.009

Koponen, A. M., Nissinen, N. M., Gissler, M., Autti-Ramo, I., Sarkola, T., & Kahila, H. (2020). Prenatal substance exposure, adverse childhood experiences and diagnosed mental and behavioral disorders - A longitudinal register-based matched cohort study in Finland. *SSM Popul Health*, *11*, 100625. https://doi.org/10.1016/j.ssmph.2020.100625

Kwon, M. K., Moore, A., Barnes, C. C., Cha, D., & Pierce, K. (2019). Typical Levels of Eye-Region Fixation in Toddlers With Autism Spectrum Disorder Across Multiple Contexts. *J Am Acad Child Adolesc Psychiatry*, *58*(10), 1004-1015. https://doi.org/10.1016/j.jaac.2018.12.011

Lambert, J. E., & Peeler, C. E. (2019). Visual and oculomotor outcomes in children with prenatal opioid exposure. *Curr Opin Ophthalmol*, *30*(6), 449-453. https://doi.org/10.1097/ICU.0000000000000604

Lieneman, C. C., Brabson, L. A., Highlander, A., Wallace, N. M., & McNeil, C. B. (2017). Parent-Child Interaction Therapy: current perspectives. *Psychol Res Behav Manag*, *10*, 239-256. https://doi.org/10.2147/PRBM.S91200

Lieneman, C. C., Quetsch, L. B., Theodorou, L. L., Newton, K. A., & McNeil, C. B. (2019). Reconceptualizing attrition in Parent-Child Interaction Therapy: "dropouts" demonstrate impressive improvements. *Psychol Res Behav Manag*, *12*, 543-555. https://doi.org/10.2147/PRBM.S207370

Liu, Y., Rodenkirch, C., Moskowitz, N., Schriver, B., & Wang, Q. (2017). Dynamic Lateralization of Pupil Dilation Evoked by Locus Coeruleus Activation Results from Sympathetic, Not Parasympathetic, Contributions. *Cell Rep*, *20*(13), 3099-3112. https://doi.org/10.1016/j.celrep.2017.08.094

Luecken, L. J., Tartaro, J., & Appelhans, B. (2004). Strategic coping responses and attention biases. *Cognitive Therapy and Research*, *28*, 23-37. https://doi.org/https://doi.org/10.1023/B:COTR.0000016928.98893.fb

McPartland, J. C., Webb, S. J., Keehn, B., & Dawson, G. (2011). Patterns of visual attention to faces and objects in autism spectrum disorder. *J Autism Dev Disord*, *41*(2), 148-157. https://doi.org/10.1007/s10803-010-1033-8

Merhar, S. L., Jiang, W., Parikh, N. A., Yin, W., Zhou, Z., Tkach, J. A., Wang, L., Kline-Fath, B. M., He, L., Braimah, A., Vannest, J., & Lin, W. (2021). Effects of prenatal opioid

exposure on functional networks in infancy. *Dev Cogn Neurosci, 51*, 100996. https://doi.org/10.1016/j.dcn.2021.100996

Merrick, M. T., Ford, D. C., Ports, K. A., & Guinn, A. S. (2018). Prevalence of Adverse Childhood Experiences From the 2011-2014 Behavioral Risk Factor Surveillance System in 23 States. *JAMA Pediatr, 172*(11), 1038-1044. https://doi.org/10.1001/jamapediatrics.2018.2537

Meyer, H. C., Sangha, S., Radley, J. J., LaLumiere, R. T., & Baratta, M. V. (2021). Environmental certainty influences the neural systems regulating responses to threat and stress. *Neurosci Biobehav Rev, 131*, 1037-1055. https://doi.org/10.1016/j.neubiorev.2021.10.014

Morie, K. P., Crowley, M. J., Mayes, L. C., & Potenza, M. N. (2019). Prenatal drug exposure from infancy through emerging adulthood: Results from neuroimaging. *Drug Alcohol Depend, 198*, 39-53. https://doi.org/10.1016/j.drugalcdep.2019.01.032

Morris, A. S., Hays-Grudo, J., Zapata, M. I., Treat, A., & Kerr, K. L. (2021). Adverse and Protective Childhood Experiences and Parenting Attitudes: the Role of Cumulative Protection in Understanding Resilience. *Advers Resil Sci, 2*(3), 181-192. https://doi.org/10.1007/s42844-021-00036-8

Morris, L. S., McCall, J. G., Charney, D. S., & Murrough, J. W. (2020). The role of the locus coeruleus in the generation of pathological anxiety. *Brain Neurosci Adv, 4*, 2398212820930321. https://doi.org/10.1177/2398212820930321

Nakakoga, S., Higashi, H., Muramatsu, J., Nakauchi, S., & Minami, T. (2020). Asymmetrical characteristics of emotional responses to pictures and sounds: Evidence from pupillometry. *PLoS One*, *15*(4), e0230775. https://doi.org/10.1371/journal.pone.0230775

Nygaard, E., Slinning, K., Moe, V., & Walhovd, K. B. (2016). Behavior and Attention Problems in Eight-Year-Old Children with Prenatal Opiate and Poly-Substance Exposure: A Longitudinal Study. *PLoS One*, *11*(6), e0158054. https://doi.org/10.1371/journal.pone.0158054

O'Brien, J. W., & Hill, S. Y. (2014). Effects of prenatal alcohol and cigarette exposure on offspring substance use in multiplex, alcohol-dependent families. *Alcohol Clin Exp Res*, *38*(12), 2952-2961. https://doi.org/10.1111/acer.12569

Oliva, M., & Anikin, A. (2018). Pupil dilation reflects the time course of emotion recognition in human vocalizations. *Sci Rep*, *8*(1), 4871. https://doi.org/10.1038/s41598-018-23265-x

Osher, D., Cantor, P., Berg, J., Steyer, L., & Rose, T. (2018). Drivers of human development: How relationships and context shape learning and development1. *Applied Developmental Science*, *24*(1), 6-36. https://doi.org/10.1080/10888691.2017.1398650

Paolozza, A., Rasmussen, C., Pei, J., Hanlon-Dearman, A., Nikkel, S. M., Andrew, G., McFarlane, A., Samdup, D., & Reynolds, J. N. (2014). Deficits in response inhibition correlate with oculomotor control in children with fetal alcohol spectrum disorder and prenatal alcohol exposure. *Behav Brain Res*, *259*, 97-105. https://doi.org/10.1016/j.bbr.2013.10.040

Radhakrishnan, R., Elsaid, N. M. H., Sadhasivam, S., Reher, T. A., Hines, A. C., Yoder, K. K., Saykin, A. J., & Wu, Y. C. (2021). Resting state functional MRI in infants with prenatal opioid exposure-a pilot study. *Neuroradiology*, *63*(4), 585-591. https://doi.org/10.1007/s00234-020-02552-3

Reh, R. K., Dias, B. G., Nelson, C. A., 3rd, Kaufer, D., Werker, J. F., Kolb, B., Levine, J. D., & Hensch, T. K. (2020). Critical period regulation across multiple timescales. *Proc Natl Acad Sci U S A, 117*(38), 23242-23251. https://doi.org/10.1073/pnas.1820836117

Richardson, G. A., Larkby, C., Goldschmidt, L., & Day, N. L. (2013). Adolescent initiation of drug use: effects of prenatal cocaine exposure. *J Am Acad Child Adolesc Psychiatry, 52*(1), 37-46. https://doi.org/10.1016/j.jaac.2012.10.011

Roberts, A. L., Koenen, K. C., Lyall, K., Robinson, E. B., & Weisskopf, M. G. (2015). Association of autistic traits in adulthood with childhood abuse, interpersonal victimization, and posttraumatic stress. *Child Abuse Negl, 45*, 135-142. https://doi.org/10.1016/j.chiabu.2015.04.010

Romens, S. E., & Pollak, S. D. (2012). Emotion regulation predicts attention bias in maltreated children at-risk for depression. *J Child Psychol Psychiatry, 53*(2), 120-127. https://doi.org/10.1111/j.1469-7610.2011.02474.x

Roos, A., Fouche, J. P., Ipser, J. C., Narr, K. L., Woods, R. P., Zar, H. J., Stein, D. J., & Donald, K. A. (2021). Structural and functional brain network alterations in prenatal alcohol exposed neonates. *Brain Imaging Behav, 15*(2), 689-699. https://doi.org/10.1007/s11682-020-00277-8

Rose, B. M., Holmbeck, G. N., Coakley, R. M., & Franks, E. A. (2004). Mediator and Moderator Effects in Developmental research. *25*(1), 58-67.

Ross, E. J., Graham, D. L., Money, K. M., & Stanwood, G. D. (2015). Developmental consequences of fetal exposure to drugs: what we know and what we still must learn. *Neuropsychopharmacology, 40*(1), 61-87. https://doi.org/10.1038/npp.2014.147

Schermerhorn, A. C., Bates, J. E., Puce, A., & Molfese, D. L. (2015). Neurophysiological Correlates of Children's Processing of Interparental Conflict Cues. *J Fam Psychol, 29*(4), 518-527. https://doi.org/10.1037/fam0000088

Schroeder, A., Slopen, N., & Mittal, M. (2020). Accumulation, Timing, and Duration of Early Childhood Adversity and Behavior Problems at Age 9. *J Clin Child Adolesc Psychol, 49*(1), 36-49. https://doi.org/10.1080/15374416.2018.1496440

Smith, K. E., & Pollak, S. D. (2020). Early life stress and development: potential mechanisms for adverse outcomes. *J Neurodev Disord, 12*(1), 34. https://doi.org/10.1186/s11689-020-09337-y

Sonon, K. E., Richardson, G. A., Cornelius, J. R., Kim, K. H., & Day, N. L. (2015). Prenatal marijuana exposure predicts marijuana use in young adulthood. *Neurotoxicol Teratol, 47*, 10-15. https://doi.org/10.1016/j.ntt.2014.11.003

Stevens, J. S., van Rooij, S. J. H., & Jovanovic, T. (2018). Developmental Contributors to Trauma Response: The Importance of Sensitive Periods, Early Environment, and Sex Differences. *Curr Top Behav Neurosci, 38*, 1-22. https://doi.org/10.1007/7854_2016_38

Syeda, M. M., & Climie, E. A. (2014). Test Review: Wechsler Preschool and Primary Scale of Intelligence–Fourth Edition. *Journal of Psychoeducational Assessment, 32*(3), 265-272. https://doi.org/10.1177/0734282913508620

Tambelli, R., Cimino, S., Cerniglia, L., & Ballarotto, G. (2015). Early maternal relational traumatic experiences and psychopathological symptoms: a longitudinal study on mother-infant and father-infant interactions. *Sci Rep, 5*, 13984. https://doi.org/10.1038/srep13984

Thomas, J. C., Letourneau, N., Bryce, C. I., Campbell, T. S., Giesbrecht, G. F., & Team, A. P. S. (2017). Biological embedding of perinatal social relationships in infant stress reactivity. *Dev Psychobiol*, *59*(4), 425-435. https://doi.org/10.1002/dev.21505

Thompson, B. L., Levitt, P., & Stanwood, G. D. (2009). Prenatal exposure to drugs: effects on brain development and implications for policy and education. *Nat Rev Neurosci*, *10*(4), 303-312. https://doi.org/10.1038/nrn2598

Thompson, R. A., & Calkins, S. D. (1996). The double edged sword: Emotional regulation. *Development and Psychopathology*, *8*(1), 163-182. https://doi.org/10.1017/S0954579400007021

Tottenham, N., Tanaka, J. W., Leon, A. C., McCarry, T., Nurse, M., Hare, T. A., Marcus, D. J., Westerlund, A., Casey, B. J., & Nelson, C. (2009). The NimStim set of facial expressions: judgments from untrained research participants. *Psychiatry Res*, *168*(3), 242-249. https://doi.org/10.1016/j.psychres.2008.05.006

Vandevivere, E., Braet, C., Bosmans, G., Mueller, S. C., & De Raedt, R. (2014). Attachment and children's biased attentional processing: evidence for the exclusion of attachment-related information. *PLoS One*, *9*(7), e103476. https://doi.org/10.1371/journal.pone.0103476

Veerapa, E., Grandgenevre, P., El Fayoumi, M., Vinnac, B., Haelewyn, O., Szaffarczyk, S., Vaiva, G., & D'Hondt, F. (2020). Attentional bias towards negative stimuli in healthy individuals and the effects of trait anxiety. *Sci Rep*, *10*(1), 11826. https://doi.org/10.1038/s41598-020-68490-5

Wass, S. V., Noreika, V., Georgieva, S., Clackson, K., Brightman, L., Nutbrown, R., Covarrubias, L. S., & Leong, V. (2018). Parental neural responsivity to infants' visual

attention: How mature brains influence immature brains during social interaction. *PLoS Biol*, *16*(12), e2006328. https://doi.org/10.1371/journal.pbio.2006328

Whiting, S. B., Wass, S. V., Green, S., & Thomas, M. S. C. (2021). Stress and Learning in Pupils: Neuroscience Evidence and its Relevance for Teachers. *Mind Brain Educ*, *15*(2), 177-188. https://doi.org/10.1111/mbe.12282

Wiggins, J. L., Mitchell, C., Hyde, L. W., & Monk, C. S. (2015). Identifying early pathways of risk and resilience: The codevelopment of internalizing and externalizing symptoms and the role of harsh parenting. *Dev Psychopathol*, *27*(4 Pt 1), 1295-1312. https://doi.org/10.1017/S0954579414001412

Wilder, L. K., & Sudweeks, R. R. (2003). Reliability ratings across studies of the BASC. *Education and Treatment of Children*, *26*(4), 382-399.

Wolicki, S. B., Bitsko, R. H., Cree, R. A., Danielson, M. L., Ko, J. Y., Warner, L., & Robinson, L. R. (2021). Mental Health of Parents and Primary Caregivers by Sex and Associated Child Health Indicators. *Adversity and Resilience Science*, *2*(2), 125-139. https://doi.org/10.1007/s42844-021-00037-7

Yeoh, S. L., Eastwood, J., Wright, I. M., Morton, R., Melhuish, E., Ward, M., & Oei, J. L. (2019). Cognitive and Motor Outcomes of Children With Prenatal Opioid Exposure: A Systematic Review and Meta-analysis. *JAMA Netw Open*, *2*(7), e197025. https://doi.org/10.1001/jamanetworkopen.2019.7025

Zahidi, R., Rogers, J., Guastaferro, W., & Whitaker, D. (2019). Relationship Between Self-Report and Observed Parenting Among Parents in Treatment Versus Not in Treatment Populations. *Journal of the Georgia Public Health Association*, *7*(2). https://doi.org/10.20429/jgpha.2019.070217

Appendix A: Stimulus Specifications

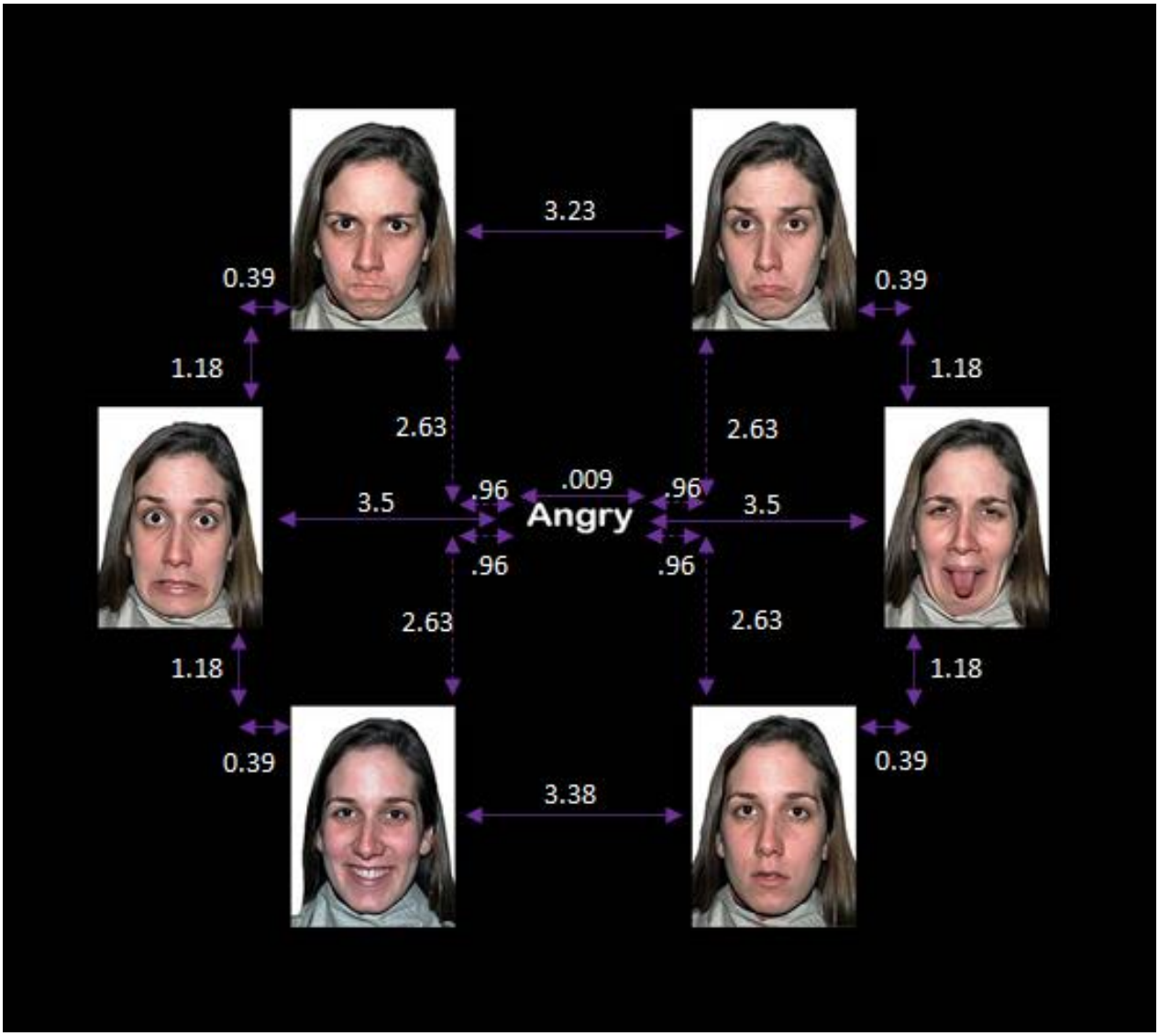

Figure A1. Adult stimulus example with dimensions. All dimensions are given in inches.

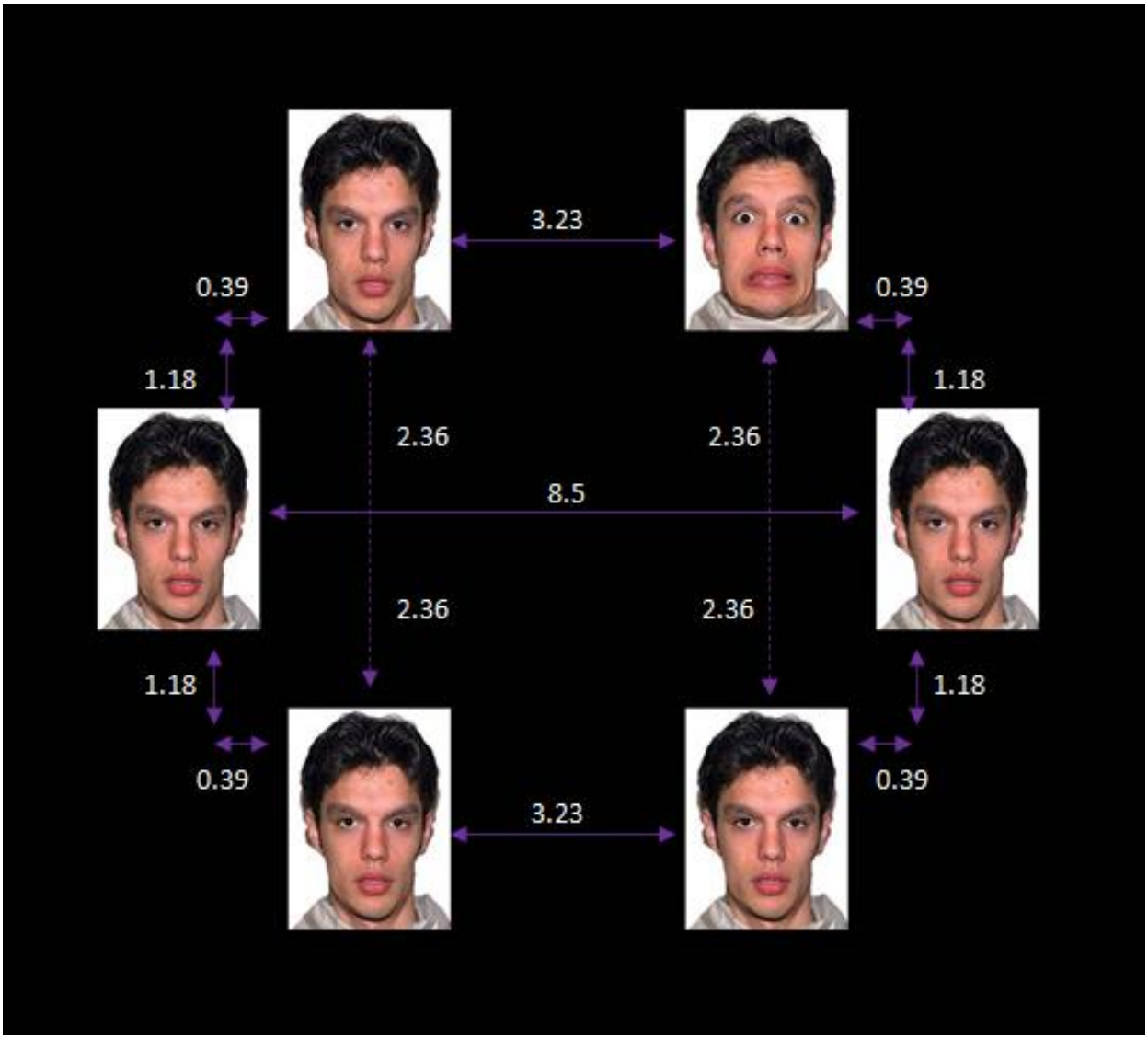

Figure A2. Child stimulus example with dimensions. All dimensions given in inches.

Appendix B: Plots Visualizing the Conditional Effect of the Focal Predictor for Primary Mediation Models

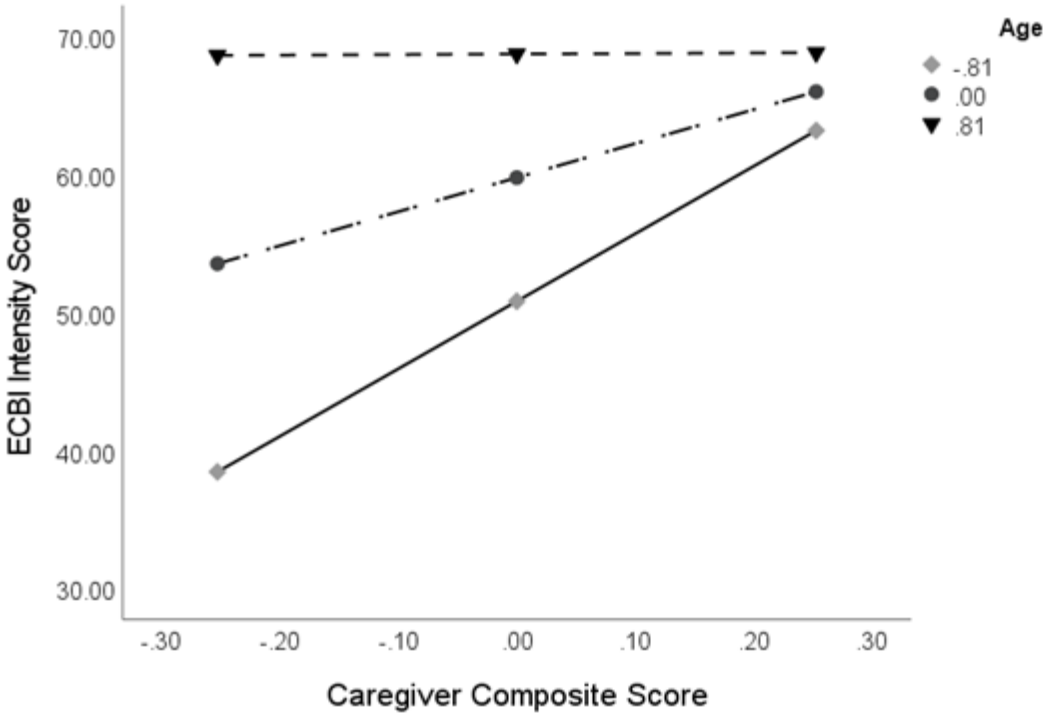

Figure B1. Data visualization for conditional effects of the focal predictor testing the relationship between general attention bias and externalizing behaviors using the ECBI intensity score.

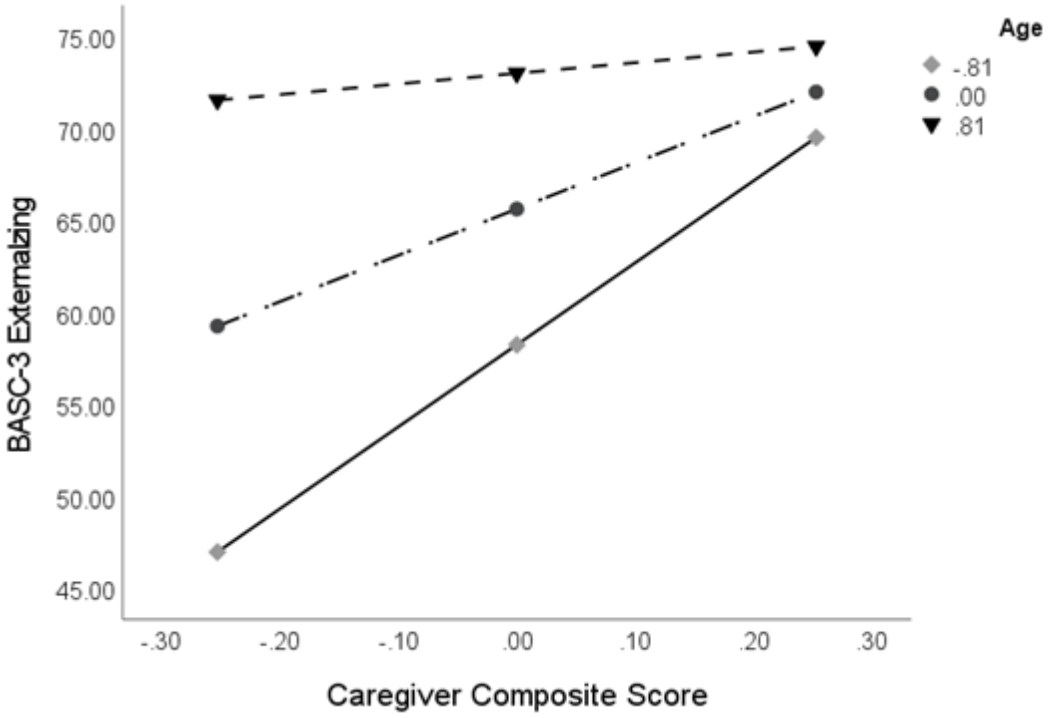

Figure B2. Data visualization for condition effects of the focal predictor for the dyad subset with ECBI scores (N = 18) testing the relationship between general attention bias and externalizing behaviors using the BASC-3 externalizing score.

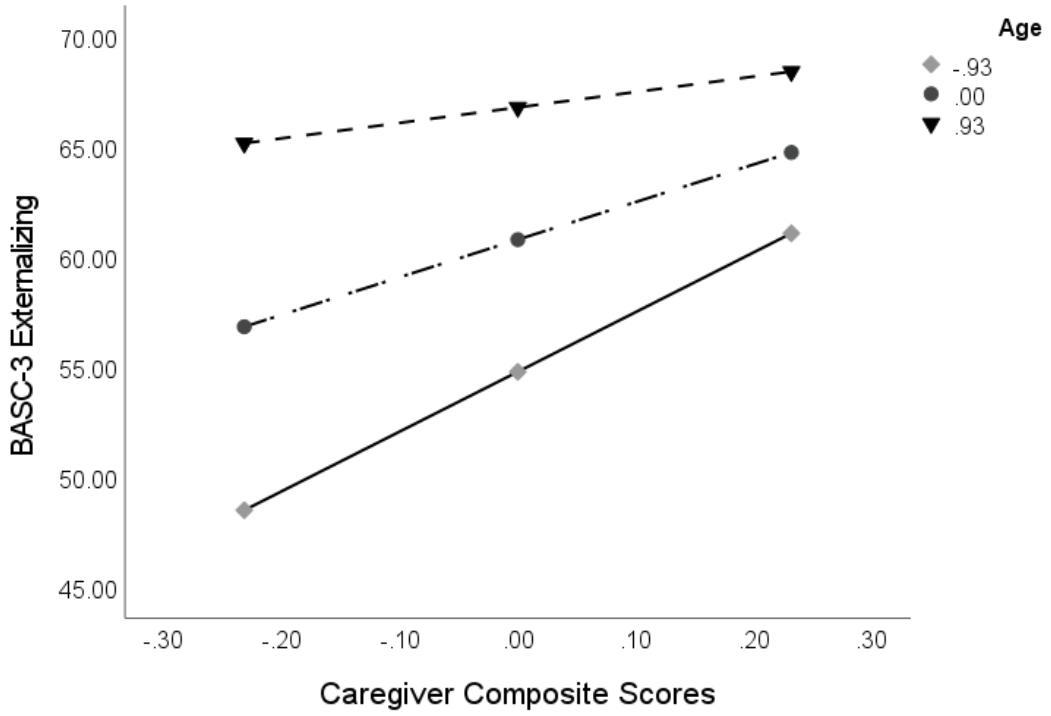

Figure B3. Data visualization for condition effects of the focal predictor testing all dyads testing the relationship between general attention bias and externalizing behaviors using the BASC-3 externalizing score.

Appendix C: Plots Visualizing the Conditional Effect of the Focal Predictor for Anger-Specific Attention Bias Mediation Models

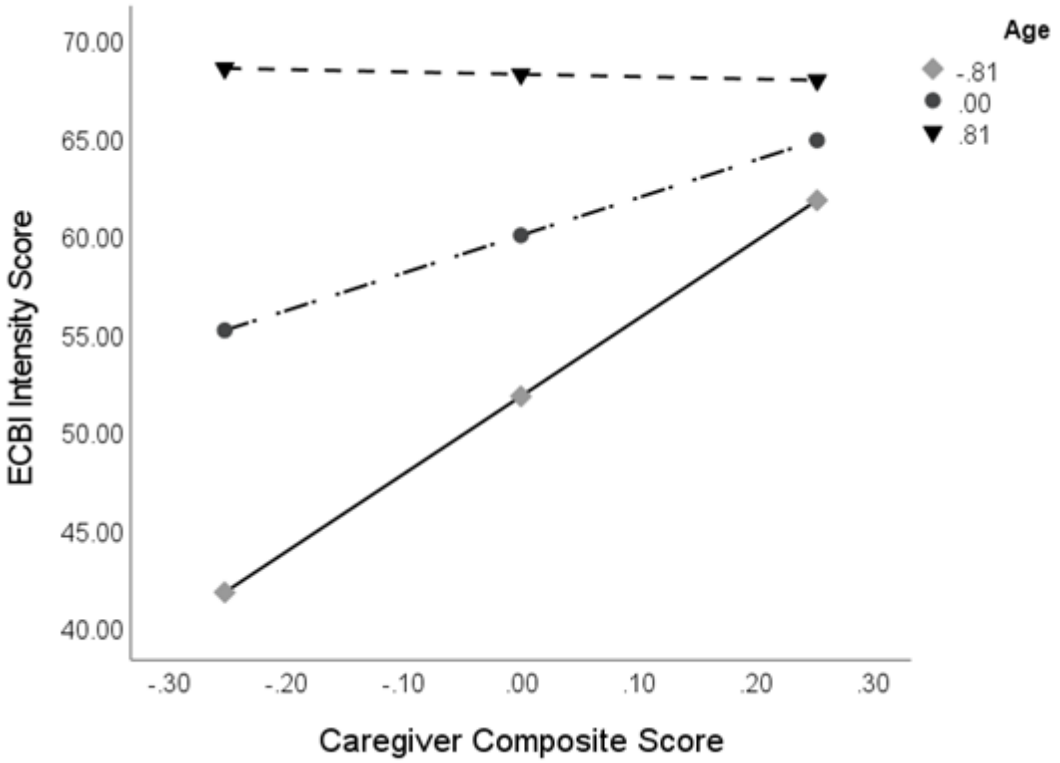

Figure C1. Data visualization for conditional effects of the focal predictor testing the relationship between anger-specific attention bias and externalizing behaviors using the ECBI intensity score.

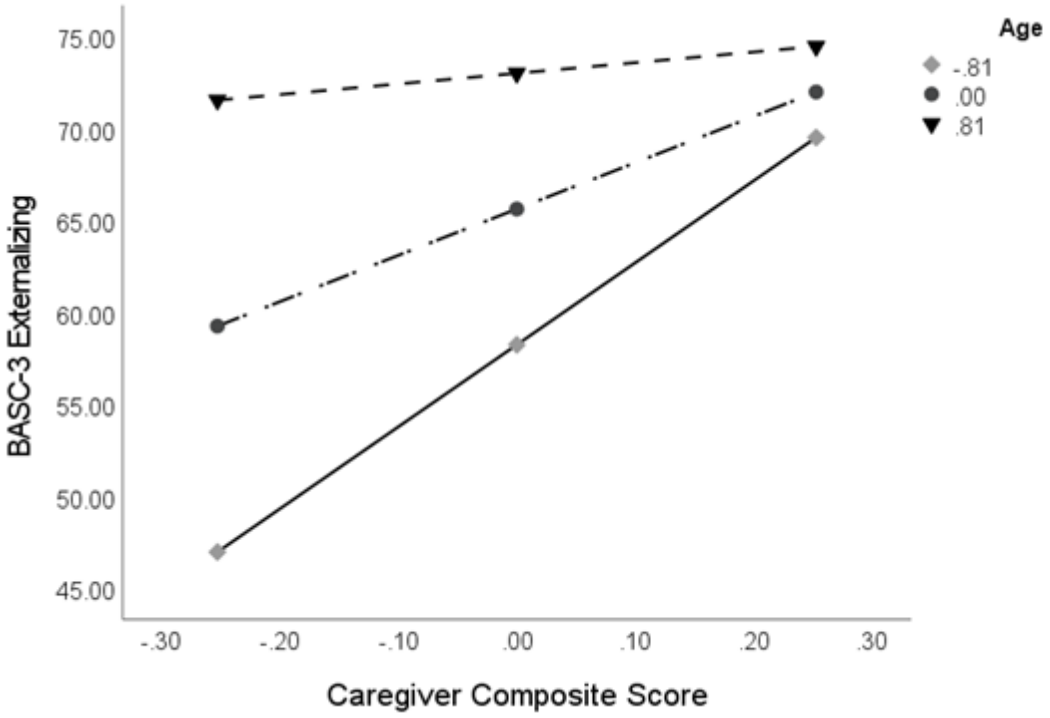

Figure C2. Data visualization for conditional effects of the focal predictor for the dyad subset with ECBI scores (N = 18) testing the relationship between anger-specific attention bias and externalizing behaviors using the BASC-3 externalizing score.

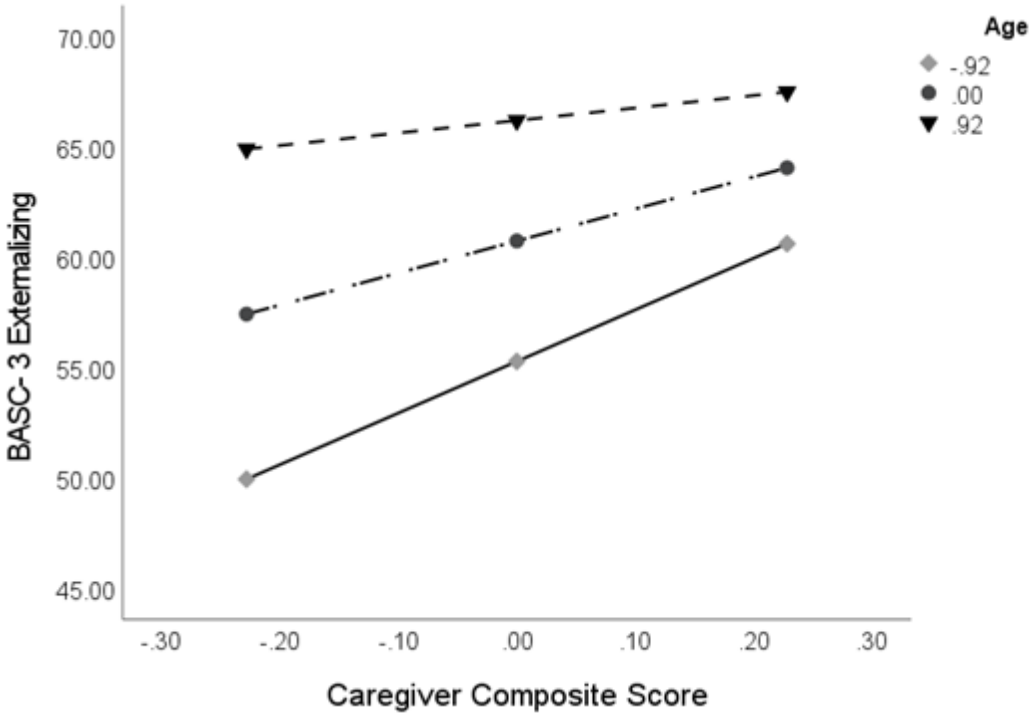

Figure C3. Data visualization for conditional effects of the focal predictor for all dyads testing the relationship between anger-specific attention bias and externalizing behaviors using the BASC-3 externalizing score.

Appendix D: Plots Visualizing the Conditional Effect of the Focal Predictor for Pupillometry Mediation Models

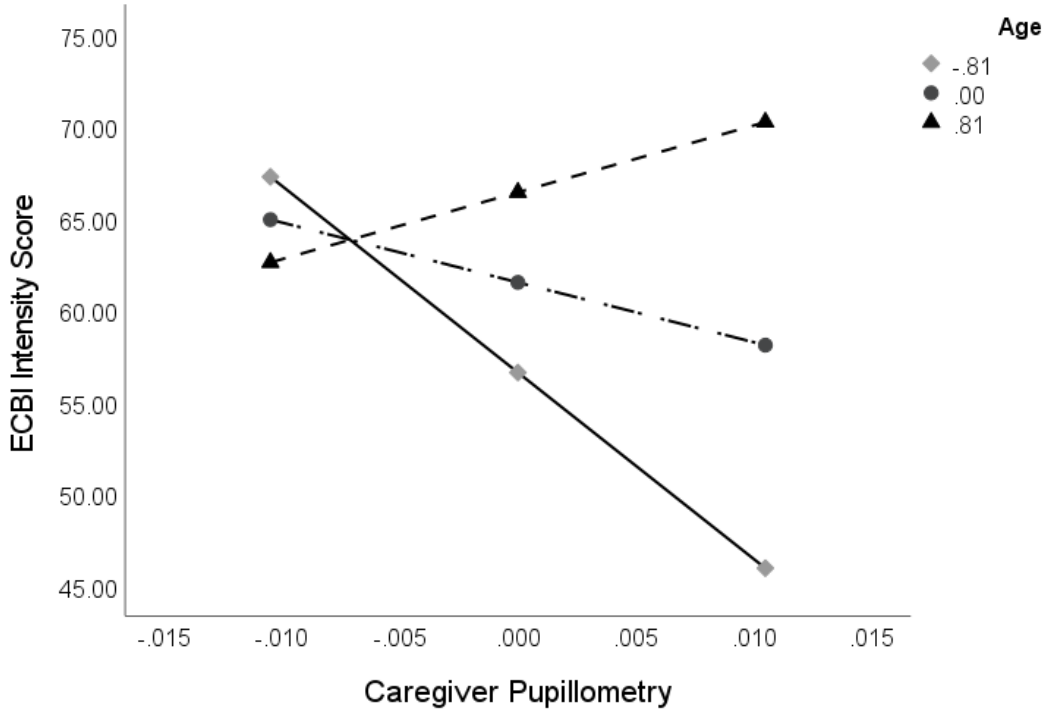

Figure D1. Data visualization for conditional effects of the focal predictor testing the relationship between caregiver pupillometry and externalizing behaviors using the ECBI intensity score.

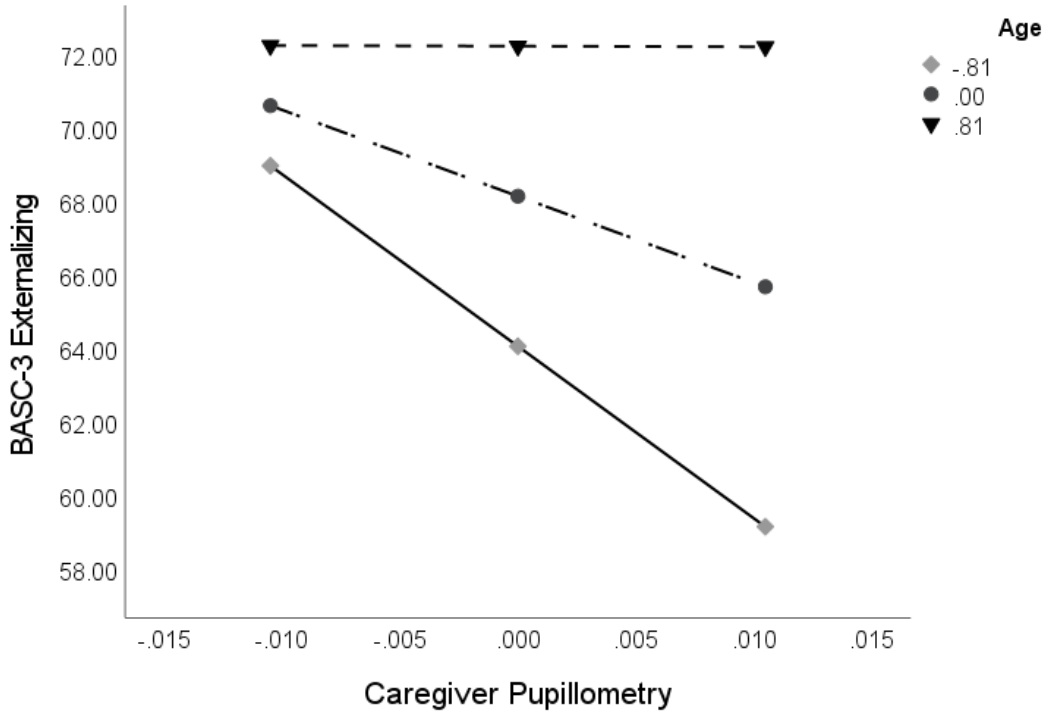

Figure D2. Data visualization for conditional effects of the focal predictor testing the relationship between caregiver pupillometry and externalizing behaviors for dyad subset using the BASC-3 externalizing scores.

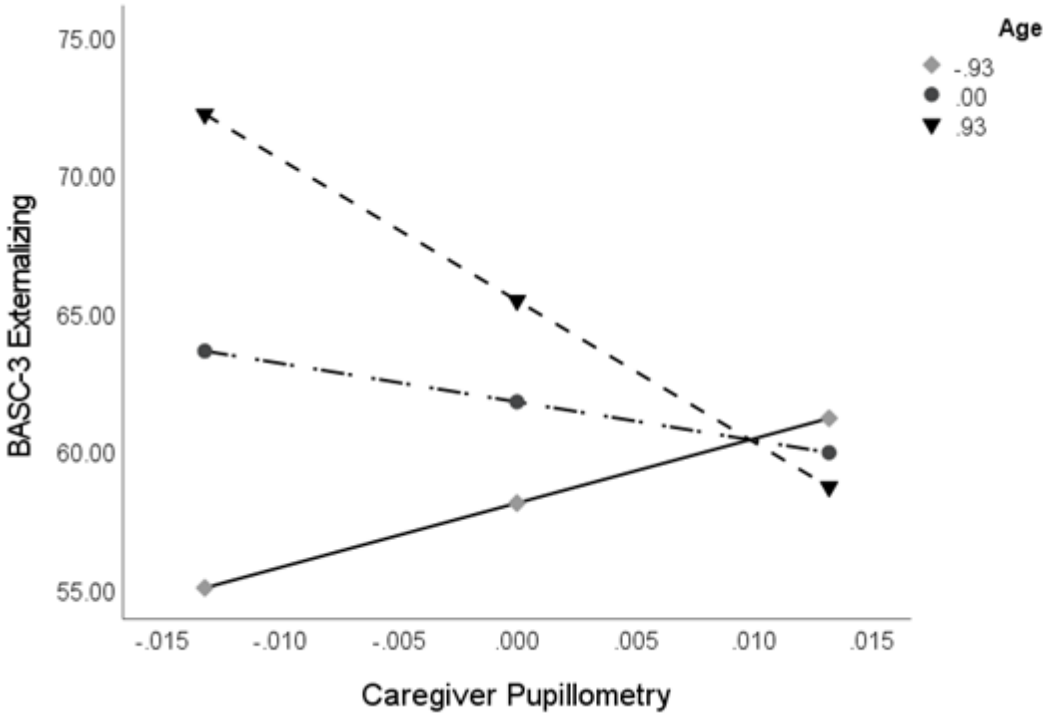

Figure D3. Data visualization for conditional effects of the focal predictor testing the relationship between caregiver pupillometry and externalizing behaviors for all dyads using the BASC-3 externalizing scores.

Appendix E: Plots Visualizing the Conditional Effect of the Focal Predictor with Emotion-Specific Caregiver Pupillometry Mediating

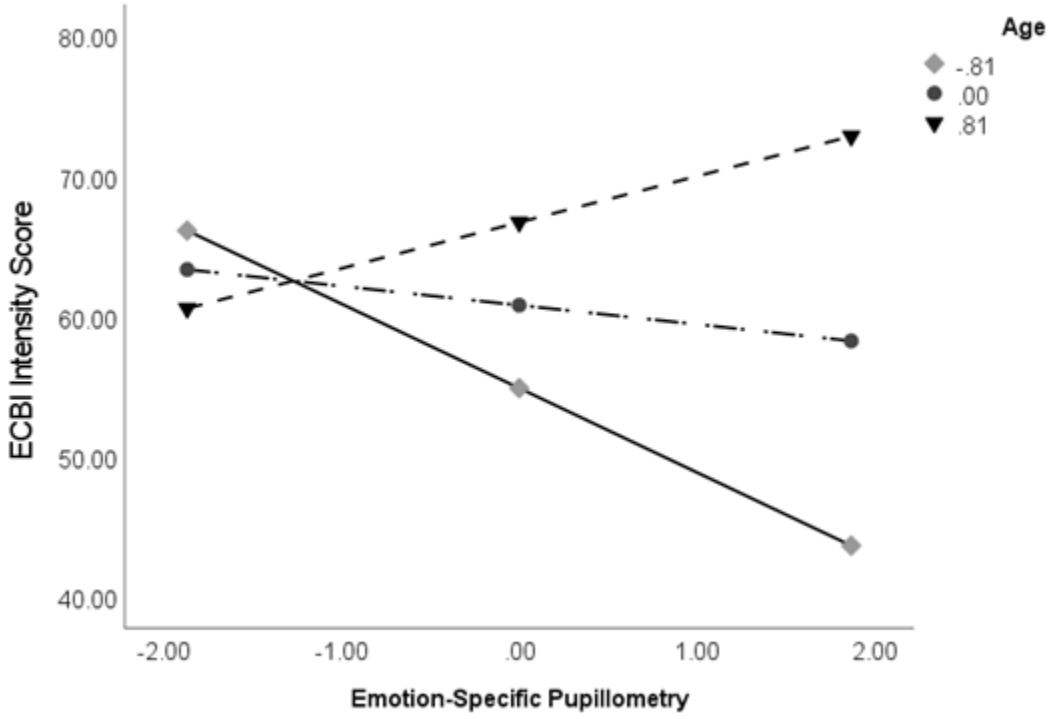

Figure E1. Data visualization for conditional effects of the focal predictor testing the relationship between caregiver pupillometry and externalizing behaviors using the ECBI internalizing scores.

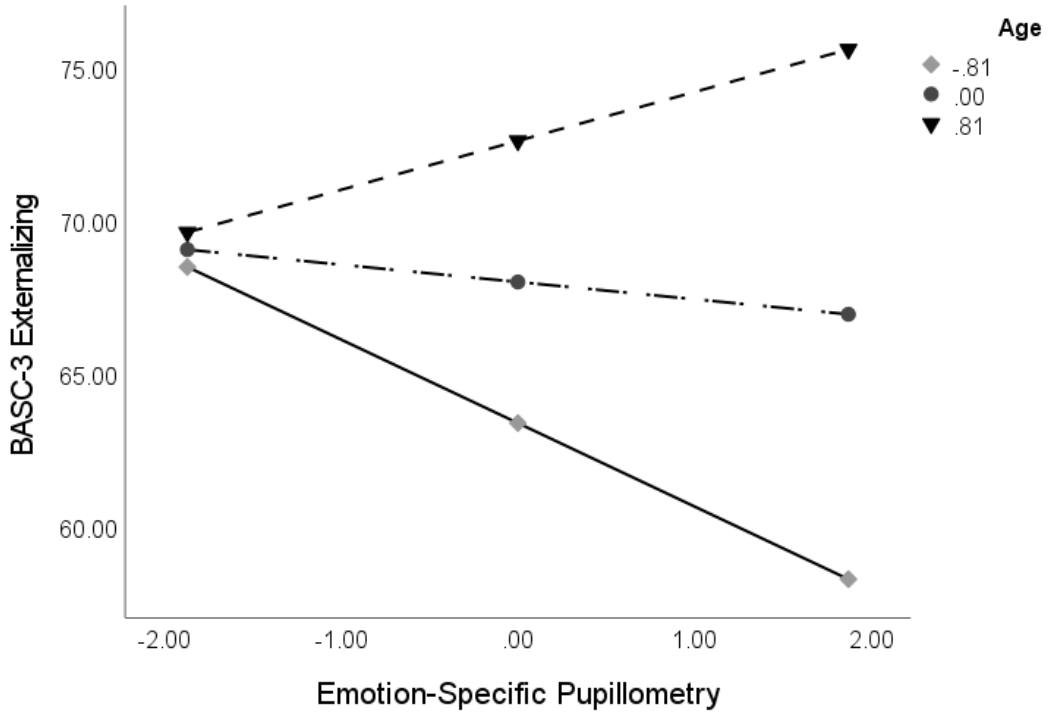

Figure E2. Data visualization for conditional effects of the focal predictor testing the relationship between caregiver pupillometry and externalizing behaviors for dyad subsets using the BASC-3 externalizing scores.

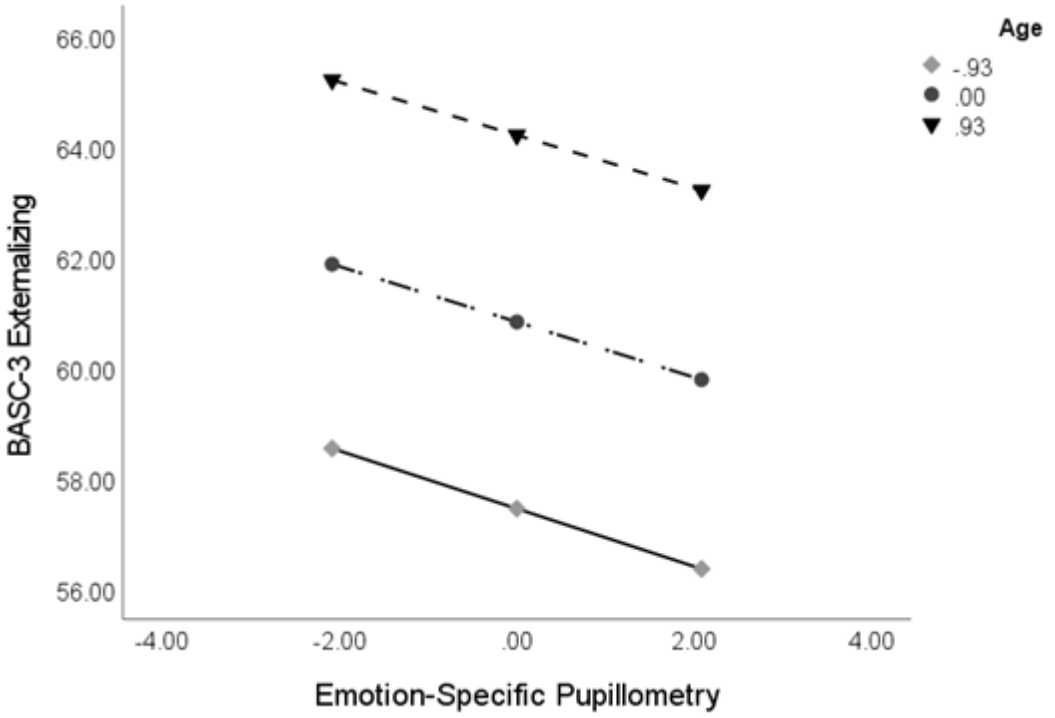

Figure E3. Data visualization for conditional effects of the focal predictor testing the relationship between caregiver pupillometry and externalizing behaviors for all dyads using the BASC-3 externalizing scores.

Appendix F: Plots Visualizing the Conditional Effect of the Focal Predictor with Caregiver Composite Mediating

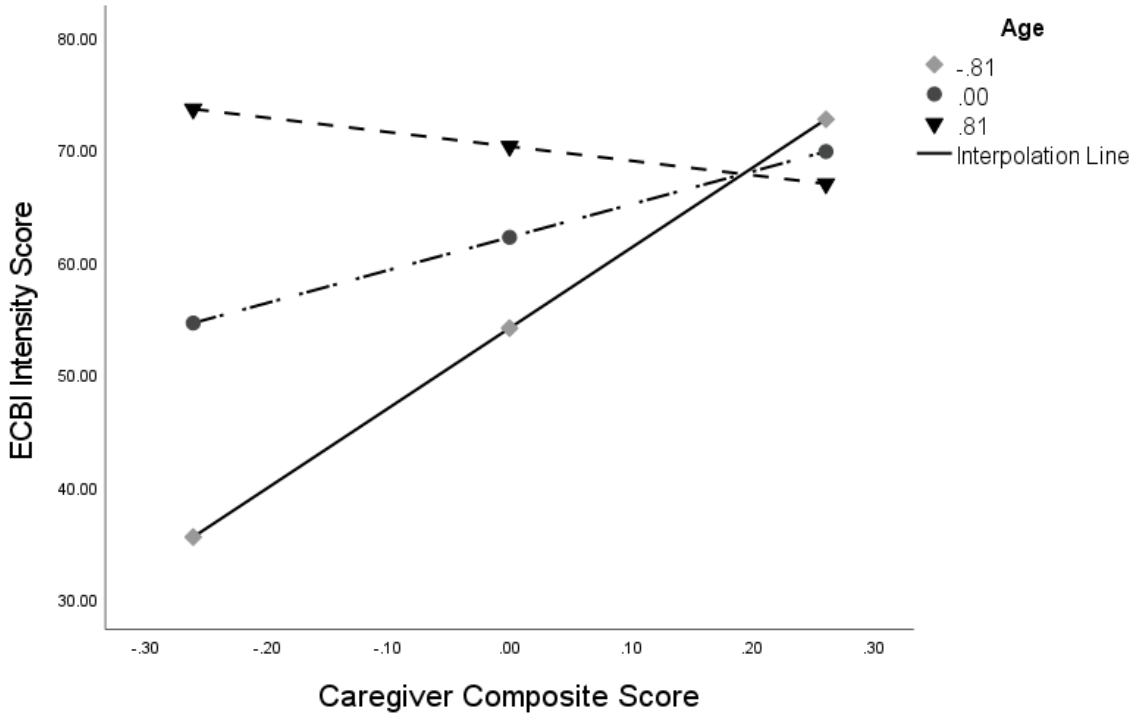

Figure F1. Data visualization for conditional effects of the focal predictor testing the relationship between caregiver composite scores and externalizing behaviors using ECBI intensity score.

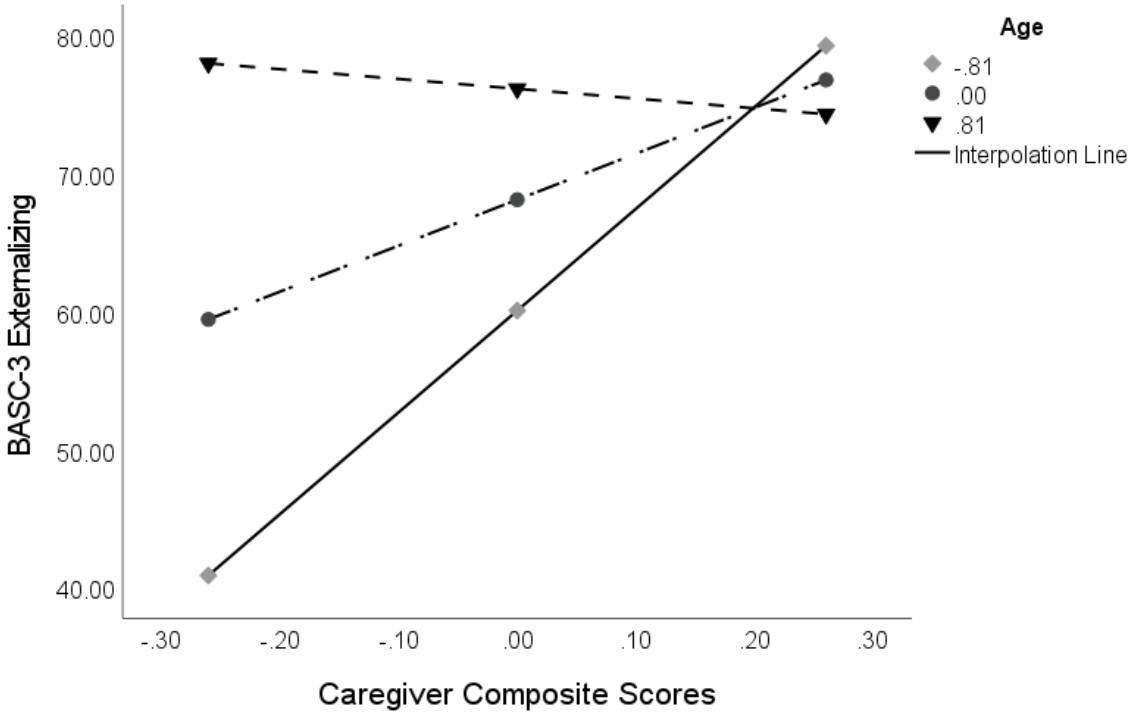

Figure F3. Data visualization for conditional effects of the focal predictor testing the relationship between caregiver composite scores and externalizing behaviors for dyad subsets using the BASC-3 externalizing score.

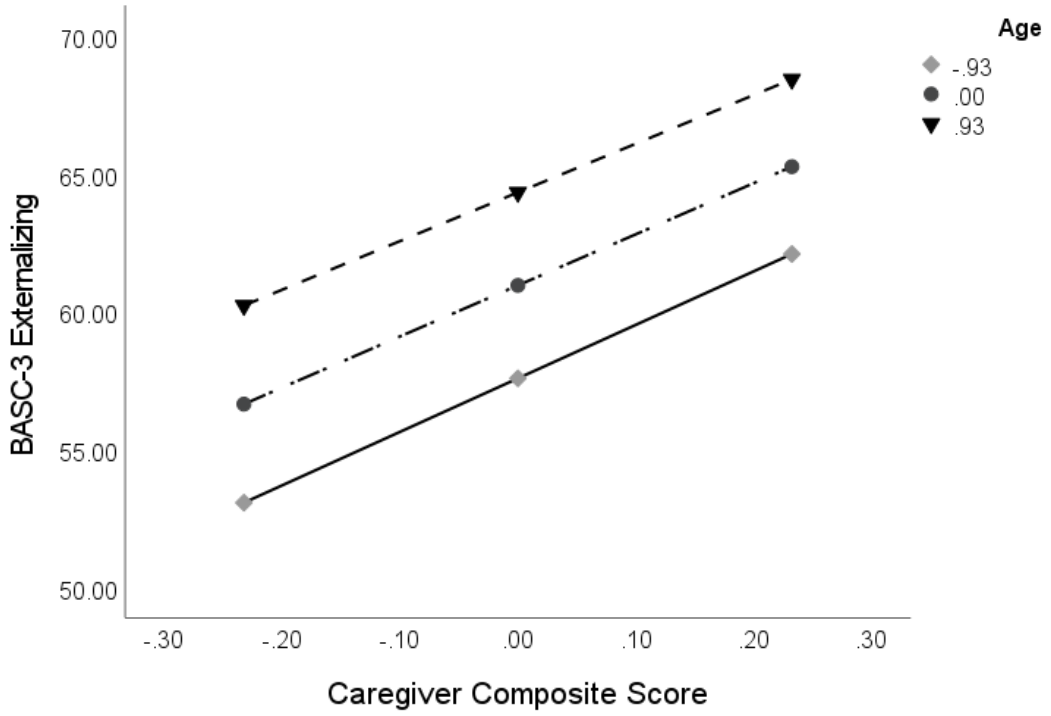

Figure F2. Data visualization for conditional effects of the focal predictor testing the relationship between caregiver composite scores and externalizing behaviors for all dyads using the BASC-3 externalizing score.