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Childhood Trauma And Emotion Processing Neurocircuitry

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To Mom, Dad, Kara, and Charlie

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CHAPTER 1: INTRODUCTION

1.1 Relevance of early emotion systems for lifelong health

Charles Darwin was one of the first to recognize that humans and some nonhuman animals are equipped with innate systems for perceiving, experiencing, and regulating emotional states (Darwin, 1897). Old as these systems are in evolution, they continue to play a critical role in regulating behavior and survival. Emotion systems are online in early life, allowing, for instance, a baby to read the facial expressions of his or her mother (Bornstein and Arterberry, 2003). Emotional expressions are considered a central form of communication for humans and other social animals (Ekman, 1992).

The range of mental health disorders characterized by emotional disturbances exemplifies the critical importance of emotion systems for human health and happiness. Emotional disturbances are, however, most central to the mood and anxiety disorders, which affect roughly 30% and 20% of the population, respectively (Kessler et al., 2005b). Considering mood disorders alone, this prevalence rate extrapolates to roughly 350 million people affected globally (World Health Organization, 2015). Emotional disorders are highly comorbid, with as many as 60% individuals with depression also having a comorbid anxiety disorder at some point in their life (Kaufman and Charney, 2000). They are also among the most debilitating and lethal conditions. Mood disorders have been named the leading cause of disability worldwide (World Health Ferrari et al., 2013, Organization, 2015), costing more than $50 billion a year in lost economic productivity (Kessler et al., 2006), $26 billion in health care (Greenberg et al., 2003), and underlie nearly 70% of all suicides (Reddy, 2010). The fact that emotional disturbances represent a chief health and societal burden is incontrovertible.

Like many other mental health conditions, mood and anxiety disorders have their roots in childhood and adolescence (Kessler et al., 2005a). There is a substantial uptick in disease onset around the time of puberty (Angold et al., 1998). Perhaps not coincidentally, this is a time when emotion processing neural systems are undergoing dramatic reorganization (Forbes et al., 2010, Pfeifer et al., 2011) and may thus be vulnerable to environmental insults or expression of genetic risk (Giedd et al., 2008). Early onset of emotional symptomology is associated with higher risk of later psychiatric morbidity, including substance use disorder, cognitive impairment, and increased risk of suicide (Birmaher et al., 2002). Suicide was named the second leading cause of death among US youth aged 12-17 (Perou et al., 2013). These findings indicate that early emotion systems set the stage for lifelong health and wellbeing. They also underscore the importance of understanding how abnormalities in emotion processing neurocircuitry in early life could lead to the development of emotional psychopathology.

Poor mental health can be detrimental to physical wellbeing. For instance, there is extensive evidence for a link between negative affect (NA), defined as the tendency to experience negative emotions (Watson and Clark, 1984), and risk of cancer, heart disease, diabetes and pulmonary disease (Spiegel and Giese-Davis, 2003, Sobel and Markov, 2005). Individuals with physical conditions, in turn, are at increased risk for cognitive and emotional problems (Turner and Noh, 1988), highlighting the bidirectional nature of brain-body health.

1.2 Brain systems that underlie emotion processing and regulation

Some of our earliest understanding of the neural bases of emotion came from lesions studies. Lesions in temporal lobes in nonhuman primates caused significant loss of emotional reactivity and aberrant social behavior (Klüver and Bucy, 1937). Lesions, for example, caused nonhuman primates to fail to exhibit their innate fear and avoidant behavior of snakes. This work

demonstrated that the medial temporal lobes are critical for emotion, which became the centerpiece for Paul MacLean's influential neuroanatomic model in 1949 (MacLean, 1949). He viewed the "limbic system" as being responsible only for emotions and represents as evolutionary older part of the brain that can be found in other mammals and reptiles. The cortex, on the other hand, is responsible for our cognitions. This view, however, turned out to be too simplistic. For instance, there are cortical sites that play critical roles in emotion processing and regulation, leading many contemporary researchers to refer to the "corticolimbic" emotion processing circuitry. Nonetheless, some of the structures identified by MacLean are still considered central emotional sites and have been the focus of much research in affective neuroscience over the past few decades.

Recent advances in functional neuroimaging have brought to light additional regions that participate in emotion processing, and also elaborated our understanding of the role that particular regions play. For instance, the amygdala was classically viewed as the *fear* center in the brain, critical for detecting and responding to environmental threats (LeDoux, 2000). More recent studies expand this view by showing that the amygdala is also engaged by positive stimuli (Fusar-Poli et al., 2009) and cues that predict reward (O'Doherty, 2004), and responds preferably to faces of a mother's own child compared to an unfamiliar child (Gobbini and Haxby, 2007). This has led to broader characterization of the amygdala as a structure important for detecting the biological, social, and emotional salience of environmental stimuli (Adolphs, 2010). The amygdala is nowadays considered a critical hub for recognizing salient information and subsequently generating appropriate reactions in physiology and behavior (Bogdan et al., 2016).

Importantly, the amygdala is a heterogeneous structure that consists of 13 cytoarchitectonically distinct nuclei (Sah et al., 2003b). These are broadly divided into three

subregions that have different functional roles and different patterns of connectivity: (1) Centromedial (CM) amygdala is involved in allocating attention to relevant stimuli (Davis and Whalen, 2001) and mediating increased vigilance (Dringenberg and Vanderwolf, 1996). (2) Basolateral (BL) amygdala has been linked to associative learning processes, and (3) Superficial (SF) amygdala has been implicated in social/affective processing (LeDoux, 2003, Phelps and LeDoux, 2005, Goossens et al., 2009). Although the spatial resolution of functional magnetic resonance imaging (fMRI) is limited, cytoarchitechtonically-based amygdala fMRI analyses have yielded consistent, replicated delineation of differential connectivity of major amygdala subregions in adults and in children (Etkin et al., 2009, Roy et al., 2009, Roy et al., 2013, Brown et al., 2014). Thus, we will test for localization of effects in different amygdala subregions.

Beyond the amygdala, additional corticolimbic emotion processing regions include the hippocampus, insula, prefrontal cortex (PFC), and anterior cingulate cortex (ACC). These regions are highly interconnected with the amygdala, and contribute to different aspects of emotional processing. The hippocampus is involved in contextual modulation of emotional expression and memory (Ji and Maren, 2007), and the insula plays a more general role in saliency processing, detecting interoceptive, autonomic and emotional information (Seeley et al., 2007). Different areas of the PFC and ACC contribute to emotion appraisal, generation, and regulation (Etkin et al., 2011). Recent reviews support a general dorsal/ventral distinction in medial PFC (mPFC) and adjacent ACC wherein dorsal areas are involved in appraisal and generation of emotion, whereas ventral regions (i.e., pregenual and subgenual regions) are involved in automatic forms of emotion regulation via suppression of amygdala activity (Etkin et al., 2011). Explicit forms of emotion regulation, in contrast, are likely subserved by dorsal PFC regions, and may work by recruiting ventral mPFC/ACC circuitry (Schiller and Delgado, 2010).

A meta-analysis of 105 fMRI emotion processing studies reveals that thalamus, striatum, brainstem, hypothalamus and visual regions also contribute to the processing of emotion (Fusar-Poli et al., 2009).

In this research, we will focus *a priori* on key regions involved in emotion processing and regulation. Specifically, we will examine the amygdala for its central role in biasing emotional processing, and amygdala-ventral anterior cingulate cortex (vACC) circuitry, which is critical for emotion regulation. The amygdala is a part of the brain's "salience network", responsible for detecting, integrating, and filtering relevant interoceptive, autonomic, and emotional information (Seeley et al., 2007, Taylor et al., 2009). In Chapters 3 and 5, we expand our view to evaluate other regions of the salience network that are highly interconnected with the amygdala, including the insula and dorsal anterior cingulate cortex (dACC). In Chapter 5 we also begin to address how childhood trauma affects interactions between salience network regions, and between the salience network and other brain networks.

1.3 Genetic and environmental variables that shift early emotion systems

We begin this section by reviewing several basic principles of brain development to provide a context for understanding typical and atypical emotion neurocircuitry in youth. The brain undergoes a protracted developmental course across the first two decades of life (Gogtay et al., 2004), mediated by an evolutionarily refined genetic program of development that coordinates the emergence of an array of behaviors at different developmental stages. Importantly, each individual holds a unique genetic plan. Thus, genetic variation between individuals can lead to variations in emotional experience and behavior, and variations in the emotion processing neurocircuitry underlying this experience.

Risk for emotional psychopathology also has a strong genetic component, evidenced by high rates of heritability and identification of common genetic polymorphisms (Cross-Disorder Group of the Psychiatric Genomics Consortium, 2013, Waszczuk et al., 2014, de Moor et al., 2015). Studies find that healthy individuals at high familial risk for depression (i.e., with an affected first-degree relative) show structural and functional variation in corticolimbic regions that is similar to affected individuals (Fusar-Poli et al., 2012, Foland-Ross et al., 2013). This suggests that at least some of the heritable risk for these disorders may be reflected by heritable variation in emotion processing neurocircuitry. Genetic factors may be particularly important for risk in early life, accounting for 40.4% of variance in risk of emotional psychopathology in adolescents (Glowinski et al., 2003).

Genetic studies implicate a large number of genes in emotional psychopathology, suggesting that heritable effects are mediated by smaller contributions of many genes. Of the most commonly studied are genes that encode for brain-derived neurotrophic factor (BDNF), the serotonin transporter (SERT), and the glucocorticoid chaperone protein (FKBP5; Mahan and Ressler, 2012). Animal research shows that these gene variants convey altered function in systems important for regulating emotional behavior and the stress response, and are associated with functional and structure variation in corticolimbic regions (Chen et al., 2006, Kalueff et al., 2010, Smoller, 2016). Examination of common functional genetic variants is appealing because of the highly conserved nature of emotion neurocircuitry across species, allowing for translation from nonhuman animal to *in vivo* human research. Indeed, studies in humans show similar effects of these gene variants on structure and function of emotion processing neurocircuitry (Hariri et al., 2002a, Mukherjee et al., 2011, Holz et al., 2015), and that many of these same gene

variants interact with environmental exposures to convey risk for emotional psychopathology (Caspi et al., 2010, Zannas and Binder, 2014).

The unfolding genetic program of brain development is modified by a simultaneous process of environmental specialization, which tailors the development to the unique experiences and environment of that individual. This occurs largely via the developmental process of pruning, which fine-tunes the synapses that are overproduced in postnatal life (Huttenlocher et al., 1982). Similar to synaptogenesis, brain regions vary in the time point at which pruning occurs. This so-called "experience-dependent plasticity" generally occurs in a sequentially and hierarchically organized manner, with the maturation of lower level functions preceding that of higher order processes (Nithianantharajah and Hannan, 2006). Each functional process, and its underlying brain circuit, undergoes a temporally limited sensitive period of heightened plasticity during which neural development is especially receptive to particular types of events. These so-called "sensitive periods" are considered evolutionarily advantageous because they 1) optimize each individual for their environment, and 2) allow for the brain to capitalize on widely available signals from the environment to scaffold development rather than relying on genetic signals alone (Bick and Nelson, 2016).

Much of what we know about sensitive periods comes from the work of Hubel and Weisel in the visual system (Hubel and Wiesel, 2004). These investigators found that binocular visual experience during early postnatal life is critical for the development of normal vision. In their experiments, they obstructed visual input to one eye very early in life in nonhuman animals. They found that visual input restored to that eye following the closure of the sensitive period failed to normalize vision. This effect was mediated via altered organization of primary visual cortex such that neurons linked to the obstructed eye were ceded to neurons encoding

information from the other eye (that received inputs). Repeating the experiment in older animals failed to show these neural and functional changes, providing evidence of a visual sensitive period. Initially, this phenomenon was conceptualized as a "critical period" rather than a "sensitive period" because the neural connections and functions become irreversible after the period closes. However, emerging research is beginning to uncover mechanisms through which plasticity can be reinstated after the closure of a sensitive period (Nabel and Morishita, 2013). This has exciting implications for mediation of aberrant developmental processes.

Although our understanding of sensitive periods stems primarily from research in sensory and motor systems, there is evidence for sensitive periods in affective development. Unlike sensory and motor systems, which develop early in life, structural and functional changes within affective neural circuits occur across the first two decades of life. Sensitive periods for affective development are perhaps most well understood during the very early postnatal period (Hartley and Lee, 2015). Work by Regina Sullivan and colleagues on amygdala fear circuitry found that prior to postnatal day 10, infant rats exhibit a paradoxical approach response to an odor previously paired with a shock. This coincides with a sensitive period for attachment learning wherein suppression of fear responding during this period may functionally promote attachment between the infant and the caregiver. Attachment therefore occurs with the caregiver, even if the quality of the care received is poor (see review by Rincon-Cortes and Sullivan, 2014). This pattern mirrors what is seen in humans: children who are abused very early in life often have strong attachment with their abusive parents (Sullivan and Lasley, 2010). After prenatal day 10 in rats, odor-shock conditioning produces an aversion, reflecting the emergence of cued fear learning. This coincides with the onset of learning-induced plasticity in the amygdala and the closure of the attachment sensitive period.

After postnatal day 10, the rat mother's presence can buffer the fear response. This suggests that caregivers play a critical role in regulating children's behavior, physiology, and stress reactivity, even after the closure of a sensitive period. Importantly, *lack* of a caregiver presence appears to cue the termination of the sensitive period for environmental input into emotion network development (Moriceau et al., 2009), resulting in early transition from approach to avoidance behavior following odor-shock conditioning (see review by Callaghan and Tottenham, 2016). These results suggest that abnormal environmental input during a sensitive period (in this case, lack of species-expected caregiver attachment) can lead to an altered developmental course, which is postulated to have negative consequences later in life (Tottenham, 2014).

There is ample evidence to suggest that lack of caregiver attachment in humans during early postnatal life can have devastating consequences. A well-studied example of this is institutional rearing of children. Orphanage-reared youth are at elevated risk for a range of negative cognitive and emotional problems, including anxiety and depression (see review by Tottenham, 2012). Research shows persistent changes in emotion processing and associated neurocircuitry, which may underlie the link between early experience and later affective dysfunction. For example, orphanage-reared youth show emotion regulation difficulties and heightened attentional biases to emotion materials (Tottenham et al., 2010). Neuroimaging studies in orphanage-reared youth show larger amygdala volume (Mehta et al., 2009, Tottenham et al., 2010, although see Sheridan et al., 2012) and altered functional connectivity (while viewing emotional face stimuli) between amygdala and mPFC (Gee et al., 2013a) - a pathway critical for emotion regulation (Kim and Whalen, 2009). Consistent with the idea of a sensitive period, these alterations persist years after the adversity ended. Indeed, many children in these

studies were adopted into families of very high socioeconomic status, most prior to age 5. In addition to the lack of a stable caregiver, the deprived environment of an institution does not provide adequate experience to scaffold normal brain development. This may be further damaging to affective development, as well as cognitive, social, sensory, and motor functioning (McLaughlin et al., 2014).

Together, it is clear that both genetic and environmental factors play a role in shaping early emotion systems. It is important to understand these factors in children and adolescents (youth), so that early indicators of premorbid risk can be identified and potentially mediated. It is likely easier to intervene during early sensitive periods, when brain systems are more malleable, than later on, when sensitive periods have closed. Closure of a sensitive period may be solidified, in part, by myelination - the final stage of brain development. Myelination occurs linearly across the first two decades of life, and then continues more slowly into middle adulthood (Westlye et al., 2010), acting as a brake on plasticity that results in circuit stabilization (McGee et al., 2005). The protracted developmental timecourse of affective systems through adolescence (Gee et al., 2013b, Gabard-Durnam et al., 2014) suggests that they have an extended period of sensitivity. This may render them more susceptible to environmental or genetic insults, but also more mutable to early remediation - a notion supported by recent data (see Bick and Nelson, 2016 for a review).

Importantly, neither genes nor environment alone are sufficient to predict the development of psychopathology. As we saw, maturity comes about via complex interactions between genetic and environmental factors over time. Thus, research on the gene-environment (GxE) interplay is increasingly emphasized for understanding both typical and atypical emotional development. In Chapter 2 and in some of our prior work (Marusak et al., 2015b), we

begin to address this by testing for a GxE effect in predicting structure and function of emotion processing neurocircuitry in youth.

1.4 Childhood trauma

As we saw in the preceding section, adverse early experiences can dramatically alter a child's emotional development. It is not surprising then, that exposure to childhood trauma (e.g., abuse, violence, neglect) is one of the most potent risk factors for an array of emotional disorders, including depression, anxiety, and posttraumatic stress disorder (PTSD; Kessler et al., 2010). This risk has been shown to persist even decades later into adulthood, accounting for an estimated 45% of child-onset and 32% of adult-onset disorders (Green et al., 2010). These findings underscore the relevance of childhood trauma for lifelong health and wellbeing. Exposure to childhood trauma is also linked to more severe forms of disease, including poorer treatment response, increased chronicity of symptoms, and heightened suicidality (Dube et al., 2001, Tunnard et al., 2014). Together, these data point to a strong and pervasive link between exposure to childhood trauma and poor mental health outcomes.

Epidemiological studies indicate that a majority of US children have been exposed to trauma by the time they reach adolescence (McLaughlin et al., 2012). Exposure to violence, crime, and/or abuse is particularly common, with rates exceeding 70% (Finkelhor et al., 2013). The alarmingly high rates of exposure and the link between childhood trauma and onset and persistence of psychopathology emphasize the need for early remediation strategies capable of circumventing the development of psychopathology. However, current therapies for emotional psychopathologies are derived from those developed for adults, despite the fact that emotion circuitry is qualitatively different in youth than in adults (Gee et al., 2013b, Gabard-Durnam et

al., 2014). Increased understanding of the neurobehavioral consequences of childhood trauma in young people is badly needed to develop empirically based, early interventions.

1.5 The functional connectome

Recognition that the genetic architecture of emotional behavior/psychopathology is complex and influenced by smaller contributions of myriad genes has prompted the investigation of biological processes that are intermediate to genes and behavioral phenotypes. Neuroimaging phenotypes of brain circuitry are lauded for their value in serving as so-called 'intermediate phenotypes', which may be more proximal to gene function and thus more accessible targets for molecular and genetic research (Meyer-Lindenberg and Weinberger, 2006, Bogdan et al., 2013). Neural circuits are also more suitable targets for clinical research because they are presumably more specific and quantifiable than diagnostic phenotypes, which are highly comorbid and heterogeneous, and thus likely collapse across several processes and circuits. For instance, under current classification systems (American Psychiatric Association, 2000, 2013), it is possible for two individuals to be diagnosed with major depression while possessing only one common symptom. Thus, in this dissertation we test the impact of childhood trauma on emotion processing neurocircuitry in youth.

Advances in neuroimaging methods, particularly functional (fMRI) and structural magnetic resonance imaging (sMRI), over the past few decades have catalyzed our understanding of affective functioning and disorders. The prevailing approach was to find specific brain region(s) linked to specific types of conditions. However, this approach has run into significant challenges; as discussed above, there are numerous distributed brain regions involved in the processing and regulation of emotions (Fusar-Poli et al., 2009). In addition, as is the case with most forms of psychopathology, neural changes in anxiety and affective disorders

are not localized to a specific brain region (Etkin and Wager, 2007). Although differences in affective disorders are most commonly observed in core emotion processing brain regions (e.g., amygdala, insula, ACC), there are also differences in interconnected regions and circuits (e.g., dorsolateral prefrontal cortex [DLPFC]).

In response to these distributed patterns, there has been increased emphasis on understanding circuit-level, and even whole brain, functional organization (Sporns, 2011). Recent advances in characterizing the "functional connectome" are due, in part, to enhanced understanding of *intrinsic* human brain organization. Research shows that the human brain is intrinsically organized into distinct functional networks that are involved in an array of emotional, cognitive, and sensory processes (Fox et al., 2005). These networks are observed even 'at rest', i.e., in the absence of a specific task, and are thought to engender and constrain brain function across an array of neuropsychological contexts. There is evidence that disruptions in specific networks contribute to specific patterns of cognitive and behavioral impairments. As such, dysconnectivity models have taken center stage in explanations of the etiology of emotional disorders and other types of psychopathologies (Greicius, 2008).

In this dissertation, we will evaluate function in localized brain regions, and begin to address how these changes relate to functional interactions within and between networks.

1.6 Current research: focus, sample, and methodological approach

The goal of this work is to identify neurobehavioral correlates of childhood trauma in youth. We focus on functional neural responses and connectivity - and to a lesser extent, structural variation - within emotion processing neurocircuitry. We will evaluate how emotional materials are processed and regulated in the brain, and also the intrinsic organization of emotion

processing circuitry at rest, when the individual is not actively engaging in emotion processing. Four inter-related studies are described, in Chapters 2-5.

Participants in this research program were recruited during the transitional developmental window of late childhood/early adolescence (ages 7-15), which marks the beginning of a period of heightened risk for the development of emotional psychopathology (Angold et al., 1998). This approach allows assessment of neurobehavioral markers prior to the emergence of a disorder but within a relatively narrow age range.

Youth participants were drawn from an urban Detroit minority community disproportionately burdened by trauma (Gillespie et al., 2009) and associated psychopathology (Lowe et al., 2014). Despite this increased apparent risk, minorities are underrepresented in clinical and neuroscientific research (Falk et al., 2013) – which has led to calls by the National Institutes of Health for greater representation of minorities in research (Brawley and Freeman, 1999). Childhood trauma was defined here as experiences of abuse (emotional, physical, sexual), violence (community, domestic) and neglect, given that these types of exposures are common and frequently co-occur (Green et al., 2010). The sociodemographic makeup and types of experiences of this population of urban youth is qualitatively different from the studies reviewed in Section 1.3 above, which focus on orphanage-reared youth adopted into higher socioeconomic status families. The studies described in each Chapter draw participants from the same overlapping sample of at-risk (i.e., lower income, minority, urban) youth. Subsamples were chosen for each Chapter based on the study aims (e.g., to match trauma and comparison groups) and available data (e.g., fMRI, salivary DNA). Study participants were run through one or two task-fMRI protocols: one consisted of an emotional face-matching task, the other, of stroop-like emotional face-categorization tasks. These tasks are described in detail, in ensuing Chapters.

The main approaches we have chosen for this research are blood-oxygen level dependent (BOLD) fMRI and functional connectivity MRI (fcMRI). fMRI uses experimental stimuli, e.g., emotional faces, to evoke neural response in brain region(s) of interest, e.g., amygdala, insula, ACC. The outcome measure is magnitude of response within localized brain regions. Because responses are quarried across the entire brain, activity in other areas can also be tested in exploratory or *post hoc* analyses.

In contrast to fMRI, fcMRI measures the *correlation* in activity/signal between different brain regions or networks. Functional connectivity (FC) reflects the underlying structural connectivity architecture of the brain, indicating the presence of direct and/or indirect (polysynaptic) connections (Greicius et al., 2009, van den Heuvel et al., 2009). FC can be measured in the context of an experimental paradigm, for instance an emotion processing task, or during rest (i.e., in the absence of an experimental paradigm). In a resting-state paradigm, individuals are told to lay awake in the scanner, and in our case, with their eyes closed. FC correlations and networks derived during the resting-state are reliable, reproducible, and linked to individual differences in temperament, behavior, and cognition (Shehzad et al., 2009, Thomason et al., 2011, Stevens and Spreng, 2014, Choe et al., 2015). With a single scan (6 min here), researchers have access to FC within a circuit of interest (e.g., FC between amygdala and mPFC/ACC) as well as patterns of whole-brain integration (i.e., within- and between-network FC).

High-resolution anatomical images are also acquired during each MRI scan session. Measures derived from sMRI include regional gray matter volume, shape analysis, and cortical thickness. In Chapter 2, we evaluate gray matter volume of the amygdala; however, multiple areas can be tested simultaneously, as done in our prior work (Marusak et al., 2015b).

In addition to neuroimaging measures, we test for effects of trauma on behavioral performance during emotion processing and regulation tasks. Presence or absence of behavioral effects informs the interpretation of neurological effects. For instance, concurrent change in brain and behavioral function might reflect a dysfunctional neurobehavioral process. This interpretation is strengthened if there is a brain-behavior correlation. Neural changes in the absence of behavioral effects, in contrast, may imply a compensatory neural mechanism (often increased neural response or FC) or a latent neural risk factor (often reduced neural response or FC). We evaluate these possibilities in our analyses.

Next, it is important to evaluate whether observed neurobehavioral changes could serve as markers of emotional psychopathology. Thus, we test for associations between neurobehavioral measures and symptoms of emotional psychopathology. As previously mentioned, emotional disorders are highly heterogeneous and consist of a range of symptoms. In recognition of this heterogeneity, there is increased emphasis on examining more fundamental underlying biological and psychological substrates. These may be observed across several disorders, thus underlying some of the phenotypic manifestations and genetic and environmental contributions shared across disorders. The National Institute of Mental Health's Research Domain Criteria (RDoC) initiative emphasizes a similar evaluation of cross-cutting mechanisms of disease (Sanislow et al., 2010, Cuthbert and Insel, 2013).

Analysis of the factors underlying mood and motivation more generally has consistently shown two distinct dimensions of affective experience: reward sensitivity (RS; other terms include behavioral activation and positive affectivity) and negativity affectivity (NA; or avoidance; Watson and Tellegen, 1985). Low RS refers to reduced motivation to seek rewards, and/or diminished positive affect in anticipation of and during receipt of rewards (Forbes and

Dahl, 2005, Forbes et al., 2009). NA is defined as the tendency to experience negative emotions, e.g., distress, fear, anxiety (Watson et al., 1985, Hasler et al., 2004). Research shows that these dimensions transcend diagnostic boundaries, with high levels of NA observed across both mood and anxiety disorders (Watson et al., 1988). This pattern may reflect the high comorbidity and genetic heritability of these conditions. Reduced RS, in contrast, is more specific to the mood disorders (and also PTSD; Glowinski et al., 2003, Kashdan et al., 2006) and is considered one of the most promising intermediate phenotypes of mood disorders (Hasler et al., 2004, Bogdan et al., 2013).

High NA and low RS are considered critical vulnerability factors in the development of emotional psychopathology, particularly during early formative years (Forbes and Dahl, 2005). NA and RS self-reported dispositions vary on a continuum of severity, even in psychiatrically healthy populations (Watson et al., 1995). As such, evaluating NA and RS dimensions as they relate to trauma-related neurobehavioral changes should improve our ability to identify specific biomarkers of risk. This approach may also inform the development of targeted interventions, as symptoms related to NA and RS respond to different types of treatments and may thus represent distinct etiopathologic mechanisms (Nutt et al., 2007).

CHAPTER 2: AMYGDALA RESPONSE TO SOCIAL CUES SHIFT WITH OXYTOCIN RECEPTOR GENOTYPE AND EARLY STRESS IN YOUTH

2.1 Introduction

In addition to its well-known role in promoting pro-social behavior, the neuropeptide oxytocin is increasingly recognized for its ability to attenuate anxiety and stress reactivity. Oxytocin administration reduces self-reported anxiety (Bartz and Hollander, 2006) and levels of cortisol (Heinrichs et al., 2003), the hormonal end product of the hypothalamic-pituitary-adrenal (HPA) axis. Thus, there is a growing interest in understanding the role of oxytocin in stress-related clinical disorders, such as anxiety, depression, and PTSD.

Variation in the oxytocin system may be an important factor in predicting risk for the development of psychiatric disorders, particularly in the context of early adversity. For example, children and adolescents exposed to early life stress (ELS), one of the most significant predictors of psychiatric illness (Green et al., 2010), exhibit lower levels of peripheral oxytocin following physical contact with their mothers (Wismer Fries et al., 2005). Further, a common variant in the oxytocin receptor gene (*OXTR*; rs2254298) has been found to interact with ELS to predict symptoms of anxiety and depression in young participants (Thompson et al., 2011). Given that sensitivity of the oxytocin system appears to be set in early life (Meinlschmidt and Heim, 2007), there is a critical need to better understand the role of oxytocin in mediating psychiatric risk during childhood and adolescence, when stress-related clinical disorders frequently emerge.

In the brain, oxytocin has marked inhibitory effects on the amygdala (Bale et al., 2001; in adult males), a corticolimbic region that plays a critical role in biasing information processing by orienting attention to salient, emotionally-laden, and biologically-relevant stimuli in the environment (LeDoux, 1998). The amygdala is also posited to govern the processing of potentially-relevant but ambiguous information in the environment, such as faces lacking

interpretable emotional content (Wright and Liu, 2006). Higher amygdala response to ambiguous faces is thought to reflect a greater tendency to perceive these cues as threatening (Forbes et al., 2011), and heightened amygdala responses to ambiguous faces are reported in individuals with anxiety (Cooney et al., 2006) and in youth at risk for depression (Dearing and Gotlib, 2009). These findings suggest that altered sensitivity of the amygdala to socially-relevant cues might contribute to the pathophysiology of these conditions (LeDoux, 1998).

Neuroimaging studies report that intranasal oxytocin administration dampens amygdala reactivity and reduces connectivity between the amygdala and brainstem regions implicated in autonomic and behavioral manifestations of fear (Kirsch et al., 2005; in adult males but not females, Domes et al., 2007). Thus, it is possible that individual variation in the oxytocin system may impact amygdala sensitivity, which may, in turn, alter the way information in the environment is processed. Consistent with this notion, adult (Inoue et al., 2010) and adolescent (Furman et al., 2011) carriers of the rs2254298 *OXTR* A-allele variant are reported to have increased amygdala volume, a neural marker thought to confer elevated emotional reactivity and anxiety (Holmes et al., 2012). These findings highlight the amygdala as an important neural substrate for *OXTR*-mediated risk for emotional psychopathology. However, it is unknown whether there is an association between oxytocin and amygdala function in early life. It is possible that increased sensitivity of the amygdala in childhood may contribute to larger amygdala volumes observed in individuals carrying the rs2254298 *OXTR* A-allele.

The present study tests the effects of the *OXTR* polymorphism rs2254298 on amygdala volume and functional responses in youth. We evaluated amygdala function using two tasks that involve processing socially-relevant face stimuli to examine the generalizability of effects across tasks. Amygdala responses to social ambiguity and during conditions of varying cognitive load

were evaluated. We predicted that A-allele carriers would show heightened amygdala responses to ambiguous social cues and a lower ability to dampen amygdala responses during higher cognitive load. We also predicted that A-allele carriers would show increased amygdala volume, as observed in prior work.

Current theory suggests that A-allele carriers may be more sensitive to environmental exposures and thus more vulnerable to the harmful effects of ELS (Brune, 2012). Emerging empirical data in youth support this notion, showing greater increases in anxiety and depressive symptoms in *OXTR* rs2254298 A-alleles than in youth with a G/G genotype following exposure to ELS (Thompson et al., 2011). Given research showing that ELS is associated with enhanced amygdala reactivity (McCrory et al., 2011; see also Chapter 4), we evaluated the hypothesis that young A-allele carriers are more sensitive to the effects of ELS on amygdala function. Here, we investigate interactive effect of ELS and *OXTR* genotype on amygdala activity. We do so in a sample of high-risk (i.e., urban, low-income, minority) youth with a high prevalence of ELS. This demographic was selected for several reasons. First, prior research shows not only that trauma frequency is more extreme in African Americans living in impoverished urban areas, but also that the negative consequences of trauma may be more severe (Alim et al., 2006). For instance, African American urban residents who experience trauma are nearly two times more likely to develop PTSD than their lower risk counterparts (Goldmann et al., 2011). Second, lower income is a significant predictor of more severe emotional psychopathology following trauma (Lowe et al., 2014). Thus, the present sample is considered high-risk due to additive effects of trauma frequency and stress burden.

2.2 Methods

2.2.1 Participants

The present study reports on 55 children and adolescents, ages 7-15, recruited through classified advertisements posted on Craigslist (Metro Detroit), printed flyers, Wayne State University (WSU) community postings, and area pediatric mental health clinics/service providers. Although 61 participants were initially included, 6 were excluded due to image artifacts that prohibited analysis of gray matter volume (GMV), as described below. Study exclusion criteria included history of neurological injury, significant learning disorder, English as a second language, or presence of magnetic resonance imaging (MRI) contraindications. Prior to the scan session, participants and parents were shown a brief video to prepare them for their MRI scan (available at: www.brainnexus.com/links). Full-Scale IQ was determined using the Kaufman Brief Intelligence Test, Second Edition (Kaufman and Kaufman, 2004b). Written informed consent and child/adolescent assent were obtained for all participants and their parents as approved by the WSU Institutional Review Board.

2.2.2 Early Life Stress and Internalizing Symptomology

ELS was measured using the 24-item Traumatic Events Screening Inventory (TESI; Ippen et al., 2002), a parent report of potential stressors experienced by the child (e.g., assault, witnessing violence, family member arrested). Number of early life stressors was calculated by summing the positively endorsed items on the TESI. Anxiety and depressive symptoms were assessed using the 41-item Screen for Child Anxiety Related Emotional Disorders (SCR; Birmaher et al., 1997) and the 10-item Children's Depression Inventory (CDI; Kovacs, 1992), respectively. A visual analog scale (VAS) was used to obtain an average rating of fear/anxiety during the MRI visit (repeat measures at 30-minute intervals) as previously described (Thomason et al., 2013).

2.2.3 Pubertal Development

Pubertal development was assessed with the self-report Tanner stages questionnaire (Marshall and Tanner, 1968). Following prior work (Forbes et al., 2011), participants were categorized as pre/early (Tanner stages 1-2) or mid/late (stages 3-5) pubertal.

2.2.4 OXTR Polymorphism Genotyping

Genetic analyses were carried out at the WSU Applied Genomics Technology Center. DNA was isolated from saliva collected in Oragene DNA collection tubes using EZ1 Advanced (Qiagen) with standard conditions. The *OXTR* polymorphism rs2254298 was investigated using a 5'-nuclease assay (Life Technologies TaqMan assay X). Assays were run in a 5 microliter reaction under standard TaqMan conditions on a QuantStudio 12K Flex (Life Technologies). Data were analyzed using TaqMan Genotyper software (version 1.3; Life Technologies).

Of the 55 total participants, 34 were carrying two G alleles (G/G homozygotes), 17 were carrying one A and one G allele (A/G), and 4 were carrying two A-alleles (A/A). Individuals heterozygous and homozygous for the A-allele were combined into an 'A-allele carrier' group (n = 21). Genetic distribution across the sample was in Hardy-Weinberg equilibrium, $\chi^2 = 1.163$, $p = 0.56$ (www.ncbi.nlm.nih.gov/snp). Power analysis based on results of a prior study of *OXTR* and amygdala GMV in youth (Furman et al., 2011) suggested that power for the current sample was 0.78.

2.2.5 MRI Data Acquisition

MRI data were acquired with a Siemens 3.0 Tesla MRI scanner (MAGNETOM Verio system, Siemens Medical Solutions) equipped with a 12-channel head coil (WSU School of Medicine MR Research Facility). fMRI data were acquired across the whole brain using a T2*-weighted echo-planar imaging (EPI) sequence with the following parameters: TR: 2000 ms, TE: 25 ms, 29 axial slices, field of view: 220 x 220 (whole brain coverage), flip angle: 90°, voxel

size: 3.44 x 3.44 x 4 mm. High-resolution anatomical images were acquired using a three-dimensional T1 magnetization-prepared rapid gradient-echo (MP-RAGE) sequence with the following parameters: TR: 1680 ms, TE: 3.51 ms, 176 axial slices, field of view: 256 x 256 (whole brain coverage), flip angle: 9°, voxel size: 0.7 × 0.7 × 1.3 mm.

2.2.6 Structural Image Processing

Prior to analysis, T1-weighted anatomical images were screened for motion artifacts (ghosting, blurring). Individual participant whole-brain intracranial masks were generated and manually corrected for N = 61 anatomical images by a trained rater (N.K.) using the interactive editing tools in the BrainSuite software package (v.13a4; Shattuck and Leahy, 2002; http://brainsuite.org/). Intrarater reliability was tested by generating whole-brain intracranial masks twice for five brains, at least two weeks apart. Intraclass correlation coefficient (ICC) was computed using the total number of voxels in each brain mask; reliability was confirmed by ICC > 0.9. Whole-brain masks and MR image volumes were then processed using BrainSuite to produce participant-specific segmentation of brain areas and derive total GMV of regions of interest (ROIs) in left and right hemispheres (see Marusak et al., 2015b for description of processing steps). The amygdala was selected as an *a priori* ROI. Prior work (Joshi et al., 2007, Joshi et al., 2009) shows that segmentation of subcortical brain structures, as performed by BrainSuite software, is highly congruent with manually segmented images drawn by experts at Massachusetts General Hospital (Dice coefficients κ= 0.6474-0.8918). Validation of subcortical segmentation in the latest version of BrainSuite is forthcoming. To ensure the accuracy of segmentation results for the current study, segmented image volumes from the automated method were manually inspected by two trained raters (N.K. and H.M.). Data from participants

showing poor quality segmentation for ROIs were excluded from corresponding analyses, as detailed below.

2.2.7 Gray Matter Volume Analyses

Total GMV of left and right amygdala ROIs was computed using BrainSuite software. Outliers that were two standard deviations above or below the mean were removed from analyses. This resulted in the removal of 1 or 2 cases per ROI. Additionally, GMV values were excluded for 3 cases per ROI due to poor quality subcortical segmentation. Upon completion of these quality assurance steps, N = 51 and 50 cases remained for structural analyses of each ROI. Results are reported for quality-assured data, and were further confirmed with the full N = 55 sample by winsorizing outliers to 2 standard deviations above or below the mean and applying whole-sample mean replacement for missing GMV values, as there was no evidence of systematic differences in those with missing values. The primary analysis was a 2x2 ANCOVA for amygdala GMV with gene group as the between-subjects factor (A-alleles carriers, G/G homozygotes) and hemisphere (left, right) as the within-subjects factor. Following prior pediatric studies (Mosconi et al., 2009), age, IQ, sex, and whole brain volume were entered as nuisance covariates. IQ has been shown to correlate with amygdala volume specifically (Rice et al., 2014) and medial temporal volume more generally (Andreasen et al., 1993, Narr et al., 2007, Amat et al., 2008). Trend-level associations were observed between right amygdala GMV and age ($r(50) = 0.27$, $p = 0.062$) and IQ ($r(41) = -0.31$, $p = 0.051$). Whole brain volume was not significantly associated with total amygdala volume, $r(49) = 0.004$, $p = 0.98$. Because *OXTR* effects previously reported were bilateral (Furman et al., 2011), we did not have *a priori* lateralization hypotheses. *OXTR*-by-sex and *OXTR*-by-age interactions were also examined. Few studies to date have evaluated oxytocin genes in African American samples, bringing this understudied

group into the wider context. Results in African American participants alone, and in follow-up analyses covarying for race, were consistent with those reported in the full sample.

2.2.8 fMRI Paradigms

Participants were run through one or two neuroimaging protocols. The same structural MP-RAGE scan was used in both protocols, but they included different fMRI tasks. The tasks involved viewing socially relevant stimuli (faces) and are widely used to probe amygdala sensitivity: (1) a Stroop-like face-categorization task (N = 29; Egner et al., 2008), and (2) a face-matching task (N = 35; Hariri et al., 2002b).

2.2.8.1 Face-Categorization Stroop Task: The Stroop task enabled us to examine the impact of high vs. low cognitive demand in the context of high arousal emotional faces. During the task (Figure 1A), participants were instructed to identify the gender of the face stimuli with a button press response, while ignoring the task-irrelevant gender word label. Face stimuli consisted of either happy or fearful expressions. Neurobehavioral responses during trials where the word label was incongruent with the face gender were compared with trials where the word label and gender were congruent. This contrast enabled us to permute cognitive demand. Prior evidence from similar Stroop (McKenna, 1986) and emotion dot probe (Armony and Dolan, 2002) studies show that stimuli with emotional valence require more attentional resources than do neutral stimuli.

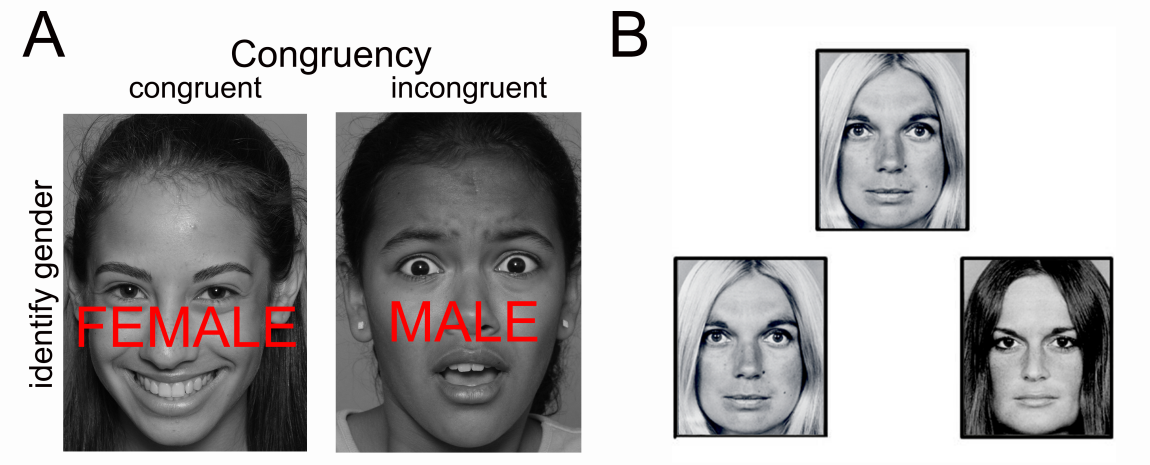

Figure 1. Experimental paradigms used to examine amygdala responses to social cues. (A) Face-categorization Stroop experimental task. Participants were instructed to indicate the gender of the face, while ignoring the overlying gender distracter word. Face stimuli included fearful and happy expressions. Trials varied such that word stimuli were either congruent or incongruent wiath the gender of the face. This enabled us to evaluate amygdala response while varying cognitive demand. (B) Face-matching task. Participants were instructed to indicate which of the two faces on the bottom of the screen matched the target face at the top of the screen. Trials were presented in emotion blocks (neutral, fearful, angry, and happy). Primary analyses focused on neutral facial expressions, given prior research showing that ambiguous faces engage variable amygdala response in youth. Gene effects on other facial expressions were also examined.

2.2.8.2 Face-Matching Task: The second task involved matching neutral faces (Figure 1B). A number of studies have shown that neutral faces are not processed as emotionally neutral, especially as perceived by youth. In fact, a still face with direct eye contact is currently the favored approach for evoking the stress response in infants (Mesman et al., 2009). Indeed, neutral faces elicit threat-related brain responses in youth (Thomas et al., 2001, Forbes et al., 2011). Amygdala response to neutral faces (particularly of adults) in children is comparable to activity elicited by negatively valenced faces in adults (Thomas et al., 2001, Lobaugh et al., 2006, Marusak et al., 2013). This observation may be due to greater perception of threat and/or increased ambiguity of neutral faces in children, as threat and ambiguity are both known to engage the amygdala (LeDoux, 1998). Thus, for children, neutral faces are less commonly used as baseline stimuli. Increasingly, abstract neutral stimuli, such as shapes, are providing more

appropriate baseline (Herba and Phillips, 2004). Here, we focused on neutral faces for their ability to elicit variable amygdala response in youth, which may reveal individual differences in negativity biases. Elliptical shapes served as the baseline sensorimotor condition. Neutral face stimuli from the most widely used Ekman face stimuli set (Ekman and Friesen, 1976) are provided in Figure S1 (see online data supplement). See (Marusak et al., 2013) for summary of face stimuli used in neuroimaging research. Effects of *OXTR* on processing non-neutral (fearful, happy, and angry expressions) were examined in follow-up analyses, given that oxytocin administration studies in adults have not consistently shown selective effects for neutral faces.

2.2.9 Functional Image Processing and Motion

During acquisition, head movements were corrected online using Siemens MRI motion correction (MoCo) software, which compares successively acquired volumes and prospectively adjusts subsequent slice acquisitions. MoCo also retrospectively corrects the time series for motion that occurred before the online correction is applied. All fMRI data reported here utilized the MoCo-corrected timeseries, and both tasks were processed using the same procedures unless otherwise noted. Data were processed with the SPM8 software package (Statistical Parametric Mapping; www.fil.ion.ucl.ac.uk/spm) implemented in MATLAB (Mathworks, Inc., Natick, Mass.). The first 4 EPI volumes were discarded to allow for system stabilization. Images were realigned to correct for motion, and the 6 motion parameters were included in participant-level models as regressors of no interest. Participants with movement exceeding 3.15 mm or 3 degrees in any direction were excluded from respective analyses (n = 9 participants total). This resulted in a final sample of 29 participants for the face-categorization task (G/G, n = 17; A-alleles, n = 12), and 35 for the face-matching task (G/G, n = 23; A-alleles, n = 12). 20 participants provided data from both tasks. For remaining participants, average movement and root-mean-square

(RMS) head position change was computed across translational (*x, y, z*) and rotational (*roll, pitch, yaw*) directions. Overall, movement was well within accepted standards (< 1.5 mm RMS; Fair et al., 2012). Movement parameters were compared between *OXTR* groups for each task using two-sample *t*-tests and a $p \leq 0.05$ (two-tailed) significance level, and reported in Results.

Images were slice-time corrected, spatially transformed to the Montreal Neurological Institute (MNI) coordinate system, and spatially smoothed with a Gaussian kernel (6 mm FWHM for the face-categorization task, 8 mm FWHM for face-matching task). Data were not resampled during normalization, thus retained the native resolution (3.44 x 3.44 x 4 mm). A 128-second temporal high-pass filter was applied, and temporal autocorrelation was estimated using a first-order autoregressive model.

Task stimuli were presented with EPrime software v.2.0 (Psychology Software Tools, Inc., Pittsburgh, PA) and displayed on a back-projection screen that was viewed by participants via a mirror attached to the head coil in the scanner.

2.2.9.1 Face-Categorization Stroop Task: The Stroop task consisted of 163 presentations of happy or fearful facial expression photographs, overlaid with the word "FEMALE" or "MALE" to create categorically congruent and incongruent stimulus sets (see Figure 2A). Participants were instructed to identify the gender of the face stimuli with a button press response, while ignoring the task-irrelevant word. Stimuli were presented for 1 s, with a varying interstimulus interval of 2-4 s (mean = 3 s) in pseudorandom order, counterbalanced across trial types for expression, word, and gender. Task duration was 12:46. Reaction time (RT) was not available for one A-allele carrier.

Participant-level models were created, as previously described (Marusak et al., 2015a). In brief, separate regressors for stimulus events were created for incongruent and congruent trials,

using 1 s boxcar functions convolved with a canonical hemodynamic response function. Error and post-error trials were modeled separately. Participant level contrasts isolated effects of conflict by contrasting incongruent - congruent trials. Average signal change was extracted for each participant from anatomically-defined left and right amygdala ROIs, using masks defined by FSL FIRST segmentation tool (Patenaude et al., 2011). A 2x2 ANOVA was used to test for *OXTR* effects on amygdala response to incongruent-congruent trials, with gene group as the between-subjects factor and hemisphere (left, right) as the within-subjects factor. Behavioral performance for incongruent vs. congruent trials was calculated and compared between groups using two-sample *t*-tests. Behavioral and neural effects were considered significant at $p \leq 0.05$ (two-tailed).

2.2.9.2 Face-Matching Task: The face-matching task consisted of 8 emotion blocks with 2 each of the following valence conditions: angry, fearful, happy, and neutral. Each block consisted of six 4 s trials, and lasted 42 s. During each trial, participants were presented with a trio of faces, and were instructed to identify which of the two faces on the bottom of the screen matched the identity of a target face on the top of the screen (Figure 2B). All stimuli in a given trial were of the same gender and face expression, and an equal number of trials depicted faces of each gender. Emotion blocks were interleaved with two sensorimotor control blocks during which participants matched the orientation of vertically- or horizontally- presented ellipses. The order of block presentation was counterbalanced across participants Task duration was 7:14. Behavioral responses for the neutral face condition were not recorded for one A-allele carrier.

Participant-level models were created, as previously described (Marusak et al., 2013). In brief, emotion blocks (neutral, fearful, angry, and happy) and baseline blocks (shapes) were modeled using convolved boxcar functions (42 s blocks). Participant level contrasts focused on

blocks of neutral face-matching vs. shapes-matching. Average signal change was extracted from anatomically-defined left and right amygdala ROIs, as described above, and compared between *OXTR* groups using a group x hemisphere ANOVA. We also tested for *OXTR* effects on behavioral performance, and on amygdala responses to other face emotions (fear, angry, happy). Analyses were re-run excluding 2 participants with less than 60% overall task accuracy, yielding no significant changes to reported results.

2.2.9.3 Localization of amygdala effects: The amygdala consists of functionally distinct nuclei that can be broadly divided into three main subdivisions: basolateral, centromedial, and superficial. We conducted follow-up exploratory analyses on voxel-wise data to test whether whole-amygdala functional effects could be localized to specific amygdala subdivision(s). Bilateral amygdala subdivision masks were generated from 3D probabilistic cytoarchitectonic maps (Amunts et al., 2005) and localization was evaluated by examining each subdivision mask for voxels surviving small-volume correction, at a family-wise error corrected threshold of $pFWE = 0.05$.

2.2.9.4 Whole-brain effects: While primary analyses focused on group difference in amygdala response, we also report results of exploratory whole-brain voxel-wise analyses at a threshold of $p < 0.005$, cluster minimum = 10 voxels.

2.2.10 Relation to Early Stress Exposure

Given prior work showing elevated levels of anxiety and depression in *OXTR* A-allele carriers in the context of ELS (Thompson et al., 2011), we tested for interactive effects of ELS and *OXTR* on amygdala volume and reactivity. Correlation analyses were first used to test for relationships between ELS and amygdala volume/reactivity, using a $p \leq 0.05$ threshold, adjusted for multiple comparisons (volume and 2 fMRI tasks: $p \leq 0.0167$). For significant effects,

PROCESS software (v.2.11; www.processmacro.org/) implemented in IBM SPSS v.21 was then used to test if ELS moderates the relationship between *OXTR* genotype and amygdala volume/reactivity.

2.3 Results

2.3.1 Participant Characteristics

OXTR groups did not differ in age, IQ, sex, income, race, pubertal development, anxiety or depressive symptoms, or whole brain volume (Table 1). *OXTR* groups also did not differ in fear/anxiety (VAS) reported during the MRI visit, $t(53) = 0.48$, $p = 0.63$. Effects observed are thus not likely influenced by group differences in stress responsivity during MRI scanning (Thomason et al., 2013). ELS was highly prevalent across the sample, with 96% reporting witnessing family threats and 32% witnessing domestic violence. Similar rates were observed within the subset of fMRI participants (see Table S2). Number of early life stressors endorsed did not differ between *OXTR* groups (Table 1). Overall, 26 participants were pre/early pubertal (Tanner stages 1-2), and 29 were mid/late pubertal (Tanner stages 3-5), $\chi^2(1) = 0.16$, $p = 0.68$. Males and females did not differ on pubertal maturation, though there was a trend for females to be more mature, $\chi^2(1) = 2.92$, $p = 0.076$.

Table 1. Demographic and Clinical Characteristics by *OXTR* Group.

Variable	OXTR G/G (n = 34)	OXTR A-Alleles (n = 21)	Group difference
Age, m (SD)	12.11 (2.42)	11.05 (2.59)	ns
Sex (Females), n (%)	23 (67.65)	11 (52.38)	ns
IQ, m (SD)	104.04 (12.33)	100.72 (16.1)	ns
Pubertal Development Group (Tanner Staging), n (%)			
Pre/Early Pubertal	14 (41.18)	12 (57.14)	ns
Mid/Late Pubertal	20 (58.82)	9 (42.86)	
Ethnicity/Race, n (%)			
African American	12 (35.3)	11 (52.3)	ns
Caucasian	12 (35.3)	6 (28.6)	

Mixed	5 (14.7)	2 (9.5)	
Hispanic	1 (2.9)	1 (4.8)	
Not reported	4 (11.8)	1 (4.8)	
Annual Household Income, n (%)			
Less than $40,000	16 (47.1)	14 (66.7)	
$40,000 - $60,000	9 (26.5)	2 (9.5)	
$60,000 - $80,000	3 (8.8)	2 (9.5)	ns
$80,000 - $100,000	2 (5.9)	0	
Over $100,000	3 (8.8)	3 (14.3)	
Not Reported	1 (2.9)	0	
Whole Brain Volume (mm^3), m (SD)	1378701.18 (140652.9)	1364428.1 (106421.75)	ns
Number of Early Life Stressors (TESI), m (SD)	2.82 (2.02)	2.05 (1.1)	ns
Anxiety Symptoms (SCR), m (SD)	16.32 (12.06)	14.95 (12.27)	ns
Depressive Symptoms (CDI), m (SD)	2.94 (3.43)	1.81 (2.79)	ns

Group difference tested using t-tests (age, IQ, anxiety), chi-square tests (sex, race), or Mann-Whitney U tests (income). Abbreviations: mean, m; not significant, ns; Intelligence quotient, IQ; Screen for Child Anxiety Related Emotional Disorders, SCR; Children's Depression Inventory, CDI: Traumatic Events Screening Inventory, TESI.

Demographics for the 29 participants in the face-categorization Stroop task and the 35 participants in the face-matching task are provided in Table S1 (see the online data supplement[1]). For both fMRI tasks, *OXTR* groups did not differ in age, IQ, income, race, pubertal development, anxiety or depressive symptoms, whole-brain volume, or movement. There was a higher proportion of female participants in the G/G homozygote group than in the A-allele group for the face-categorization task, $\chi^2(1) = 5.15$, $p = 0.033$. Follow-up analyses controlling for sex yielded no changes to results reported.

2.3.2 Amygdala Volume

ANCOVA controlling for age, IQ, sex, and whole brain volume revealed a main effect of *OXTR* on amygdala volume, $F(1, 44) = 4.15$, $p = 0.048$, $\eta_p^2 = 0.086$. As predicted, this effect

[1]http://www.sciencedirect.com.proxy.lib.wayne.edu/science/article/pii/S0028393215301913

was driven by higher GMV in A-alleles compared to G/G homozygotes (see Figure 2A). The main effect of hemisphere, main effect of sex, and hemisphere x *OXTR* interaction were not significant, *ps* > 0.2. The main effect of *OXTR* on amygdala GMV remained significant when all covariates except IQ were removed, $F(1,37) = 5.32$, $p = 0.027$, and became $F(1,46) = 3.85$, $p = 0.056$ when all except age were removed.

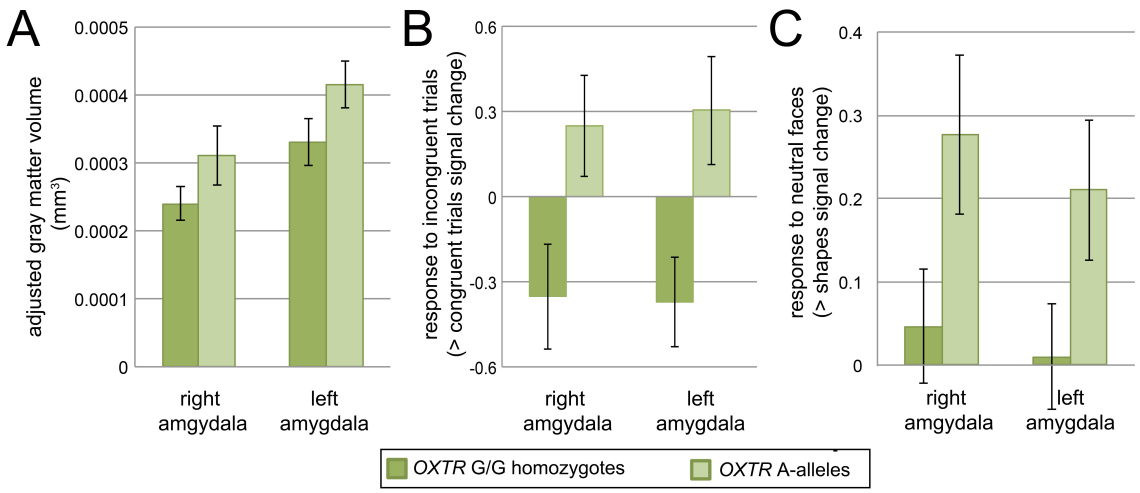

Figure 2. *OXTR* A-alleles show increased amygdala gray matter volume (GMV) and response when viewing socially-relevant face cues. GMV values are adjusted for whole brain volume. Error bars represent standard error of the mean. Main effect of *OXTR, ps* < 0.05.

2.3.3 Amygdala Reactivity

2.3.3.1 Face-Categorization Stroop Task: Consistent with previous work, reaction time (RT) across the sample was slower for incongruent (mean = 876.3 ms, SD = 232.17 ms) relative to congruent (mean = 842 ms, SD = 211.35 ms) trials, $t(27) = 3.67$, $p = 0.001$. Similarly, accuracy was lower for incongruent (mean = 84.67%, SD = 12.17%) than congruent (mean = 88%, SD = 9.67%) trials, $t(28) = 2.89$, $p = 0.007$. *OXTR* groups did not differ in their degree of RT interference, $t(26) = 0.9$, $p = 0.37$. Compared to G/G homozygotes, A-alleles showed a greater reduction in accuracy for incongruent relative to congruent trials, $t(27) = 2.33$, $p = 0.027$

(see Table 2 for behavioral performance, by group). This behavioral impairment was accompanied by higher average signal in the amygdala in *OXTR* A-allele carriers during incongruent relative to congruent trials (Figure 2B), $F(1, 27) = 7.01$, $p = 0.013$. There was no main effect of hemisphere or hemisphere x *OXTR* interaction, $Fs < 0.1$, $ps > 0.6$. The pattern of increased amygdala response and impaired behavioral performance is consistent with greater interference by incongruent stimuli in A-allele carriers. *Post hoc* within-group hemisphere x trial-type (congruent, incongruent; beta weights) ANOVAs revealed a main effect of trial-type in G/G homozygotes, $F(1,16) = 5.08$, $p = 0.039$, but not in A-allele carriers, $F(1,11) = 2.49$, $p = 0.14$. That is, amygdala activity was lower during incongruent relative to congruent trials in G/G homozygotes but not in A-allele carriers. This suggests that the observed higher amygdala response in A-allele carriers was driven by a lower ability to dampen amygdala response during incongruent trials. Evaluation of trial types separately (incongruent, congruent) showed no differences between groups ($ps > 0.05$). Magnitude of amygdala response was not correlated with any measure of accuracy or RT across participants, or within either gene group (all $ps > 0.05$).

Table 2. Performance for face-categorization and face-matching tasks, by *OXTR* group.

Face-categorization conflict task	**OXTR G/G (n = 17)**	**OXTR A-Alleles (n = 12)[1]**	***p***
Accuracy (%), congruent trials, m (SD)	85.12 (9.9)	92 (8.1)	ns
Accuracy (%), incongruent trials, m (SD)	83.82 (11.3)	85.83 (13.74)	ns
RT (ms), congruent trials, m (SD)	831.87 (203.37)	857.69 (232.34)	ns
RT (ms), incongruent trials, m (SD)	873.04 (223.9)	881.34 (240.75)	ns
Face-matching conflict task	**OXTR G/G (n = 23)[2]**	**OXTR A-Alleles (n = 12)[3]**	***p***
Accuracy (%), task overall, m (SD)	95.15 (14.26)	93.82 (13.45)	ns
Accuracy (%), neutral face condition, m (SD)	94.36 (21.34)	97.58 (8.37)	ns
RT (ms), task overall, m (SD)	1114.31 (233.76)	1159.03 (216.22)	ns
RT (ms), neutral face condition m (SD)	1105.24 (314.19)	1171.13 (232.96)	ns

Abbreviations: reaction time, RT. [1]RT data unrecorded for one A allele. [2]Behavioral data not recorded for one G/G participant. [3]Behavioral data not recorded during the neutral face condition for one A-allele participant.

2.3.3.2 Face-Matching Task: Neither accuracy nor RT differed between *OXTR* groups for neutral face-matching, matching other face valences (fearful, angry, happy), for the shapes-matching condition, or for neutral vs. shapes (all $ts < 1.3$, all $ps > 0.2$; see Table 2 for behavioral performance by group). Neuroimaging data showed that amygdala response to neutral faces was higher in A-allele carriers than G/G homozygotes, $F(1,33) = 4.14$, $p = 0.05$ (Figure 2C). No main effect of hemisphere or hemisphere x *OXTR* interaction was observed, $ps > 0.18$. The effect of *OXTR* was not driven by group differences in amygdala activity during the baseline shapes condition (vs. implicit baseline), $F(1, 33) = 0.14$, $p = 0.7$. To test whether effects of *OXTR* genotype were specific to ambiguous faces, we conducted a 3-way (hemisphere x emotion x gene-group) ANOVA to examine amygdala responses to other face emotions, i.e., fearful, happy, and angry. No main effects or interactions were observed ($ps > 0.2$). Amygdala response was not correlated with accuracy or RT during the neutral face condition across the sample (accuracy: $r(34) = -0.19$, $p = 0.3$; RT: $r(34) = -0.15$, $p = 0.43$) or within either group ($ps > 0.16$).

2.3.3.3 Localization of functional responses in the amygdala: Follow-up exploratory analyses on voxel-wise data evaluated whether group differences in whole-amygdala response could be localized to specific subregion(s). For the face-categorization conflict task, *OXTR* group differences were significant in basolateral amygdala, $pFWE = 0.011$, $Z_{max} = 3.63$, $x = -30$, $y = -4$, $z = -34$ (MNI). For the face-matching task, *OXTR* effects on neutral face-matching could not be unequivocally localized to a specific subregion, *pFWEs* 0.054-0.063.

2.3.3.4 Whole-brain results: Results of exploratory whole-brain fMRI analyses are provided in Table S4 (see online data supplement). Briefly, A-allele carriers showed higher activation to conflict (incongruent-congruent trials) than G/G homozygotes in posterior insula, amygdala, hippocampus, midbrain, and brainstem. There were no regions in which activity to

conflict was elevated in G/G homozygotes relative to A-alleles. For neutral face-matching, A-alleles showed elevated response in amygdala, posterior insula, parahippocampal gyrus, middle temporal gyrus, and inferior frontal gyrus. G/G homozygotes had greater activity in striatum, superior temporal gyrus, and inferior parietal lobe.

2.3.4 Relation to Early Stress Exposure

2.3.4.1 Amygdala volume: Neither left nor right amygdala volume was related to number of early stressors in either gene group or across the sample, $rs < |0.15|$, $ps > 0.6$.

2.3.4.2 Amygdala reactivity: During the face-categorization task, we found a positive association between right amygdala response and number of early life stressors in A-allele carriers, $r(11) = 0.705$, $p = 0.015$ (Figure 3). The correlation remained significant if nonparametric tests were applied, $r_s(11) = 0.749$, $p = 0.008$. The association between ELS and amygdala response was not significant for left amygdala, $r(11) = 0.32$, $p = 0.34$, or in G/G homozygotes (left: $r(22) = -0.08$, $p = 0.71$; right: $r(22) = 0.06$, $p = 0.79$). Correlation between right amygdala response during the face-categorization task and ELS was significantly higher among A-allele carriers than among G/G homozygotes, $Z = 1.94$, $p = 0.05$. Moderation analyses confirmed that ELS moderates the relationship between *OXTR* genotype and right amygdala reactivity, $\Delta R^2 = 0.36$, $\Delta F(1,24) = 3.93$, $p = 0.02$, $b = 0.48$, $t(24) = 2.46$, $p = 0.02$. ELS was not correlated with amygdala response to neutral faces across the sample (left: $r(33) = -0.13$, $p = 0.47$; right: $r(33) = -0.25$, $p = 0.15$) or in either gene group ($ps > 0.4$).

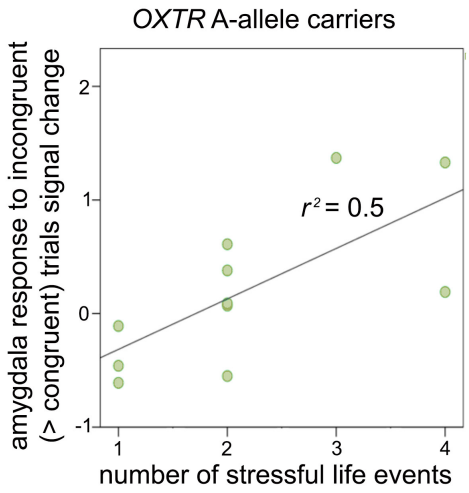

Figure 3. Relation between amygdala reactivity and early life stressors (ELS) present in *OXTR* A-allele carriers but not G/G homozygotes.

2.4 Discussion

Emerging data suggest that genetic variation in *OXTR* modulates emotion processing and regulation, and thus likely plays a role in susceptibility to psychopathology. The present study demonstrates novel links among *OXTR* genotype, ELS exposure, and structure and function of the amygdala in early life. We report that amygdala GMV and responses to social cues shift with oxytocin receptor gene variant rs2254298 in a sample of high-risk children and adolescents. Specifically, compared to youth with the G/G genotype, youth carrying the A-allele showed increased volume and higher response of the amygdala across two tasks that required viewing socially-relevant face cues. Increased amygdala responses in A-allele carriers corresponded to poorer task performance during the face-categorization Stroop task. The link between *OXTR* genotype and amygdala reactivity was moderated by ELS exposure, such that the magnitude of amygdala response was associated with ELS in A-allele carriers but not G/G homozygotes. These findings provide empirical support for recent hypotheses that *OXTR* rs2254298 A-allele carriers are more susceptible to the deleterious effects of adverse environmental exposures compared to their G/G counterparts (Brune, 2012).

Consistent with prior findings in adults (Inoue et al., 2010) and adolescents (Furman et al., 2011), we observed increased amygdala GMV in *OXTR* rs2254298 A-allele carriers. We augment these prior studies by demonstrating that amygdala function is also altered in youth carrying the A-allele. Given the amygdala's critical role in detecting the salience, significance, ambiguity, and personal relevance of environmental stimuli (LeDoux, 1998), higher amygdala responsivity may reflect greater biological tuning to socially-relevant information. While increased attentional capture of relevant social information, such as faces, may be adaptive for survival, it may also interfere with cognitive function if task demands are high. In particular, because processing capacity of the brain is limited, emotion processing must be dampened to effectively carry out higher-order cognitive processes. For instance, regional blood flow of the amygdala decreases during cognitively demanding tasks, whereas it increases during specific emotion-related tasks (Drevets and Raichle, 1998). In line with this framework, we observed that higher amygdala response, likely reflecting emotional processing, led to impaired task performance in A-allele carriers when cognitive demands were high (i.e., during the face-categorization Stroop task). In contrast, when tasks demands were low (i.e., during the face-matching task), elevated amygdala activity observed in A-allele carriers did not predict lower performance. This dissociation is consistent with conceptual models proposing that emotion processing can hinder or facilitate ongoing processing depending on the level of cognitive demand (Pessoa, 2009).

Follow-up voxel-wise analyses indicated that *OXTR* effects on amygdala response during the face-categorization task were likely localized to the basolateral amygdala subdivision. Basolateral amygdala is a main target of sensory afferents to the amygdala (McDonald, 1998) and plays an important role in social cue processing (Adolphs, 2001). Receiving highly

processed sensory information enables basolateral amygdala to detect biologically relevant stimuli in the environment (Sah et al., 2003a). Importantly, administration of intranasal oxytocin appears to alter activity in the basolateral amygdala (Gamer et al., 2010), which may partially explain oxytocin's implicated role in attenuating fear responses. In this light, lower ability to dampen basolateral amygdala response to conflict may reflect lower filtering of sensory information in *OXTR* A-allele carriers. Consistent with this hypothesis, elevated neural activity was accompanied with greater performance decrements to conflict in A-alleles.

Prior neuroimaging studies show that oxytocin administration dampens amygdala responses to salient social cues (Domes et al., 2007; in adult males). By altering oxytocin levels or receptor binding/density, the *OXTR* polymorphism may similarly modulate the tuning of the amygdala to salient social information. Here, we found *OXTR* effects on amygdala activity during face processing in youth. We did not observe *OXTR* effects during processing of fearful, angry, or happy expressions, but found response to neutral faces to differ by *OXTR* genotype. This suggests that, for children, the *OXTR* polymorphism affects sensitivity to ambiguous social threat cues, but not to signals with clearer biological relevance (e.g., an angry face signals direct threat to the perceiver). As noted above (see *Methods)*, elevated amygdala response to facial expressions of neutral valence may correspond to a negative interpretation of these stimuli in youth. Future studies will be needed to determine if the observed increase in amygdala response in A-alleles coincides with a greater perception of threat, negativity, or ambiguity in these cues.

Heightened perceptual processing of ambiguous social cues may increase risk for disorder in youth who are predisposed to interpret these cues as threatening, such as those reared in adverse environments. In accord with this notion, we discovered an association between amygdala reactivity and ELS exposure in *OXTR* A-allele carriers. Youth with ELS carrying the

OXTR A-allele appeared to manifest increased correlates of risk for internalizing conditions; they had greater amygdala responses to salient social cues. It is possible that those with both genetic susceptibility and ELS exposure not only exhibit increased sensitivity to conflicting or ambiguous stimuli, but also process them more deeply, thereby increasing susceptibility to negative rumination. This interpretation is consistent with research indicating that youth at risk for depression have a higher tendency to perceive ambiguous stimuli as threatening (Dearing and Gotlib, 2009), and fits with cognitive theories of depression which hold that greater tendency to interpret ambiguous situations as negative precipitates negative mood states (Foland-Ross and Gotlib, 2012). And indeed, elevated anxiety and depressive symptoms are reported in *OXTR* A-alleles in the context of ELS (Thompson et al., 2011).

Given that impairments in social behavior are ubiquitous in neuropsychiatric disorders, the observed effects of the *OXTR* polymorphism on amygdala processing of social cues have relevance for disease. These findings may also have implications for adolescent development outside the realm of psychopathology. In particular, higher amygdala reactivity to ambiguous social stimuli in *OXTR* A-allele carriers could hamper development in the social domain, as these individuals may be less likely to engage in social risk-taking and exploration or to initiate new peer relationships. On the other hand, such a behavioral pattern might also serve a protective function by increasing wariness even in the absence of overt threat cues. These predictions are supported by research showing that intranasal oxytocin administration in humans increases willingness to accept social risks arising through interpersonal interactions (Kosfeld et al., 2005), as well as animal literature highlighting oxytocin's effects on pro-social approach behavior (Lukas et al., 2011).

To our knowledge, this is the first study to examine neural variation associated with the *OXTR* polymorphism rs2254298 in a predominantly African American sample. By replicating effects observed in other demographic samples (Inoue et al., 2010, Furman et al., 2011) this work extends findings to wider ethnic populations. We found that amygdala structural and functional neurobiology varies with rs2254298 genotype in youth. Although mixed-race samples may improve generalizability of findings, results may be biased by population substructure and allelic frequencies may differ by population. The current analysis was sensitive to these concerns. First, allele frequencies in the sample were in line with expected frequencies for the racial majority group (African Americans). Second, although genomic control or ancestry informative marker methods were unavailable here, we did attempt to rule out population stratification by reanalyzing the data controlling for self-reported race/ethnicity, which shows high correspondence with genotyped markers (Divers et al., 2011, Levran et al., 2012). Additionally, *OXTR* groups did not differ on race, and sub-analyses in African American participants (the racial majority) were consistent with results reported in the full sample. Findings of these control analyses are consistent with a lack of confounding population substructure. Nonetheless, our sample of urban, low-income, minority youth represents an under-studied population at high risk for ELS and associated adverse outcomes (e.g., depression, PTSD, substance use disorders). Thus, research in this area may be useful for informing broader public health policies and practices. These results should nevertheless be considered preliminary and replication in other samples is needed.

We note several limitations of the current study. First, the functionality of the *OXTR* polymorphism rs2254298 is currently unknown. While higher peripheral oxytocin levels have been reported in *OXTR* rs2254298 A-allele carriers compared to G/G homozygotes (Apter-Levy

et al., 2013), how this variant translates into the actual availability or binding of oxytocin in the brain is unclear. Next, prior studies in adults using acute oxytocin administration have identified sex differences (Domes et al., 2010, Weisman et al., 2013) that were not replicated here. This may be the result of immature sexual dimorphism in childhood, because this was a gene variant rather than an acute administration study, or because this interaction may have been underpowered. These possibilities cannot be resolved here, but lack of sex differences have been observed in adult studies that evaluated the gene variant investigated here (Inoue et al., 2010). This introduces the idea that acute effects of oxytocin administration differ from more stable effects of functional gene polymorphisms. In addition, sex distribution did not differ between groups for volumetric analyses or for the face-matching task. For the face-categorization task, there was a higher proportion of female participants in the G/G homozygote group relative to the A-allele carrier group. However, a converging pattern of results across tasks, namely higher amygdala reactivity in A-alleles, suggests that observed effects are not driven by sex. Nonetheless, our power to detect *OXTR*-by-sex interactions was limited and we urge future studies with larger samples to address this important question. Third, rs2254298 is one of two widely studied oxytocin-system related polymorphisms. We did not examine effects of other common *OXTR* variants, which may also modulate oxytocin system function. Here, we chose to focus on rs2254298 to extend previous work documenting effects of this polymorphism on amygdala volume and emotional symptomology in a separate sample of mixed-race youth (Thompson et al., 2011). A recent meta-analysis (Bakermans-Kranenburg and van IJzendoorn, 2014) found null effects of both *OXTR* rs2254298 and *OXTR* rs53576 on psychopathology and social behavior. The current study, as well as prior work in adolescents (Thompson et al., 2011) demonstrate that genotype alone may not be sufficient to produce psychopathology. Rather, a

stress diathesis model may be more appropriate for understanding the downstream consequences of genetic variation in the *OXTR* gene.

Given what appears to be a critical developmental period for solidifying the relation between genotype and amygdala structure and function in the context of ELS, regulation of amygdala sensitivity may be a promising target for preventive interventions in early life before morphological changes become hardwired in the brain or before clinically-significant conditions emerge. For instance, selective serotonin reuptake inhibitors, often used to treat anxiety and depression, are effective at attenuating amygdala responses. Likewise, oxytocin administration has been shown to dampen amygdala responses, and is emerging as a potential therapeutic for stress-related clinical conditions. Given evidence that amygdala responsivity is under control of both oxytocin and serotonin (Mottolese et al., 2014), interventions should take into account individual variation in both systems.

CHAPTER 3: ALTERED AMYGDALA CONNECTIVITY IN TRAUMA-EXPOSED URBAN YOUTH

3.1 Introduction

Estimates regarding rates of exposure to traumatic events in childhood range widely from 15-60% (Kessler et al., 1995, Dube et al., 2001, Stein et al., 2010), with strong evidence for the highest incidence occurring in urban, inner city settings (almost 90%; Gillespie et al., 2009, Goldmann et al., 2011). Trauma and stress injure the brain, precipitate cognitive-behavioral, emotional, and somatic problems, and are strong predictors of psychiatric illness (McEwen, 2012). Trauma in early life is especially harmful – associated with about 50% of childhood psychiatric disorders, and 30% of later-onset clinical disorders (Green et al., 2010).

Neurological evaluation of the link between trauma and psychiatric illness converges on amygdala and prefrontal brain regions. The amygdala is essential for the detection of threat and enhancement of vigilance (Zald, 2003), and is a central activator of the physiologic stress response (Dedovic et al., 2009). In contrast, the prefrontal cortex (PFC) is critical for regulation of emotion (Quirk and Beer, 2006). Animals that experience early life adversity show structural and functional changes in both the amygdala (see Malter Cohen et al., 2013b for a review) and PFC (review by McEwen and Morrison, 2013). Furthermore, early life adversity may also perturb the direct bidirectional connections between these two regions (review by Tottenham and Sheridan, 2009).

Research in humans and animals consistently demonstrates altered frontoamygdala *connectivity* is another consequence of early life stress (Gee et al., 2013a, Malter Cohen et al., 2013a, Philip et al., 2013, Grant et al., 2014). Compromised connectivity between the amygdala and PFC has been implicated in the pathophysiology of stress-related disorders. Specifically, altered frontoamygdala connectivity has been observed in anxiety (Kim and Whalen, 2009, Roy

et al., 2013), depression (Tang et al., 2013), and PTSD (Edwards et al., 2013, Stevens et al., 2013, Brown et al., 2014). It is possible that altered frontoamygdala connectivity may emerge early in life, proximal to negative traumatic experiences, and that this shift may be formative in determining healthy or deleterious outcomes for the individual. After all, frontoamygdala circuitry undergoes rapid changes across childhood and adolescence (Hare et al., 2008, Gee et al., 2013b, Gabard-Durnam et al., 2014), and thus alterations occurring during this period may have lasting effects.

While animal research has addressed embedding of stress exposure in frontoamygdala pathways in early life, research in humans is relatively recent. Gee (2013a) and Nooner (2013) provided the first studies of early life stress and functional connectivity (FC) in children/adolescents. Gee and colleagues examined task-related variation in FC associated with orphanage rearing in 6 to 17 year olds during a face-processing task, whereas Nooner and colleagues examined how trauma-symptoms in a healthy sample of community youth relate to variations in intrinsic FC during resting-state. Both studies evidenced altered frontoamygdala FC, with orphanage-reared children showing more medial frontal and typically developing youth showing more lateral frontal amygdala FC effects. Frontoamygdala FC alterations have also been shown to relate to trauma, diurnal cortisol, and internalizing symptoms in a longitudinal community sample of young adults (18 year olds; Burghy et al., 2012, Herringa et al., 2013a). These studies compel the need for more research during formative years, prior to when psychopathology becomes chronic. Research that examines youth at high risk for developing clinical disorders could lead to identification of latent neural risk factors contributing to psychopathology in the aftermath of trauma exposure.

Here, we examine intrinsic resting-state neural FC in urban, low-income, minority trauma-exposed and comparison youth (ages 9-15). Past research shows not only that trauma frequency is extreme in African Americans living in impoverished urban areas, but also that negative consequences of trauma in urban, African American communities may be more severe (Alim et al., 2006). For example, while approximately 20% of trauma-exposed individuals in the general population subsequently develop PTSD, African American urban residents who experience trauma are nearly two times more likely develop PTSD (Goldmann et al., 2011). In addition, lower income is a significant predictor of more severe emotional psychopathology following trauma (Lowe et al., 2014). Thus, additive effects of trauma frequency and stress burden may be particularly deleterious to healthy emotional development. Investigating the correlates of trauma exposure on the developing brain in a low income, African American urban cohort of youth provides an opportunity to identify neurological changes in those who are at highest risk for developing psychopathology.

3.2 Methods

3.2.1 Participants

The current study evaluated 42 urban youth, ages 9-15 (mean = 12.6, SD = 2.1). Participant ages were selected to align with the emergence of puberty; puberty has been identified as a time when affective disorders frequently manifest (Angold et al., 1998). The majority of participants (n = 27) reported annual incomes of less than $40,000, and only a small number (n = 6) reported incomes greater than $60,000. Participants were drawn from a larger study, and chosen to represent trauma-exposed (n = 21) and comparison (n = 21) groups matched on age and sex. Data from 18 participants from both groups have been reported previously (Thomason et al., 2013). Participants were recruited through advertisements posted on the

Wayne State University website, Craigslist (Detroit), printed flyers, or through Metro Detroit mental health clinics. Exclusionary criteria included: English as a second language, lower than a 2nd grade reading level, history of brain injury, neurological or movement disorders, or presence of magnetic resonance imaging (MRI) contraindication. Parental informed written consent and child/adolescent assent were obtained prior to participation. Demographic and behavioral measures were administered during a laboratory visit, and MR imaging was performed during a subsequent visit (~2 weeks following the laboratory visit). All experimental procedures were approved by the Human Investigation Committee of Wayne State University.

3.2.2 Demographic data analysis

Highest level of parent/caregiver educational attainment and annual household income were coded as ordinal variables and compared between groups using Mann-Whitney U tests. Independent samples *t*-tests or chi-square tests were used to test for group differences in age, sex, IQ (derived from the Kaufman Brief Intelligence Test, version 2; Kaufman and Kaufman, 2004b), parent report of child race/ethnicity, and pubertal development (using Tanner staging; Marshall and Tanner, 1968). Effects were considered significant at $p \leq 0.05$. Statistical analyses were 2-tailed, and implemented in IBM SPSS Statistics 21 (SPSS, Inc., Chicago, IL).

3.2.3 Trauma exposure

Utilizing both parent and child endorsements, participants that experienced at least one trauma indicated on the Children's Trauma Assessment Center Screen Checklist were categorized as 'trauma' (source: Michigan Trauma Assessment Center). Forms of trauma included significant events that threatened their safety (e.g., physical or sexual abuse), undermined their security (e.g., neglect), caused unmanageable stress (e.g., witness a violent

crime), or compromised interpersonal and family relationships (e.g., emotional abuse). Number and type of endorsed traumas are provided in Table 3.

3.2.4 Self-reported affect measures

Participants completed two validated self-report questionnaires: the 10-item Children's Depression Inventory (CDI; Saylor et al., 1984) and the 41-item Screen for Childhood Anxiety-Related Disorders (SCR; Birmaher et al., 1997). The SCR was administered during the lab visit and again at the MRI visit. Total SCR score was summed for each visit and then averaged, as lab and MRI scores were highly correlated, $r(42) = 0.76$, $p < 0.001$. A visual analog scale (VAS) was used to obtain an average rating of fear/anxiety during the MRI visit (repeat measures at 30-minute intervals) as previously described (Thomason et al., 2013).

3.2.5 Imaging data acquisition

MRI data were acquired using a 3.0 Tesla Siemens Verio scanner. Participants were positioned in a 12-channel head coil and stabilized by padding to reduce motion-related artifacts. Participants were asked to lie quietly in the scanner with their eyes closed for the duration of the six-minute resting-state scan. For fMRI, a total of 180 T2*-weighted BOLD images were acquired (interleaved ascending acquisition) using EPI. The acquisition parameters were: repetition time [TR] = 2000 ms; echo time [TE] = 25 ms; flip angle = 90°; voxel size= 3.44 x 3.44 x 4 mm; matrix = 220 x 220; and 29 slices. Additionally, T1-weighted images were obtained for anatomical reference with the following parameters: time [TR] = 1680 ms; echo time [TE] = 3.51 ms; flip angle = 9°; voxel size = 0.7 x 0.7 x 1.3 mm; matrix = 256 x 256; and 176 slices.

3.2.6 Image preprocessing

Image preprocessing steps were conducted using SPM8 software (Statistical Parametric Mapping; http://www.fil.ion.ucl.ac.uk/spm/). After discarding the first 4 EPI volumes to allow for signal stabilization, images were slice-time corrected, realigned, spatially normalized to the Montreal Neurological Institute (MNI) template, and smoothed using an 8-mm Gaussian kernel. Frame-to-frame excursion, root mean square (rms), and head movement across the scan were calculated and averaged for translational (x, y, z) and rotational (roll, pitch, yaw) movement directions. Motion parameters were compared between groups using 2-tailed independent samples *t*-test. Between group motion differences were considered significant at $p < 0.05$.

3.2.7 Seed-based connectivity analysis

Given prior research showing altered intrinsic FC of specific amygdala subregions in adults with PTSD (Brown et al., 2014) and anxiety (Etkin et al., 2009), connectivity of centromedial (CM) and basolateral (BL) amygdala were evaluated separately. Amygdala subregions were the leading choice for seeded ROI analyses because of their unique connectivity profiles. For more detailed discussion of amygdala subregion connectivity in humans, see (Roy et al., 2009). The present study was centered on CM and BL subregions as these as these have previously been implicated in stress-related psychiatric disorders (Etkin et al., 2009, Brown et al., 2014). Seed regions were defined for CM and BL amygdala structural subdivisions (Amunts et al., 2005), following prior work (Roy et al., 2009, Qin et al., 2012). In brief, bilateral masks used stereotaxic, probabilistic maps of cytoarchitectonic boundaries defined by the SPM Anatomy toolbox (Eickhoff et al., 2005). Cytoarchitectonic maps show high reliability and accuracy for guiding anatomical segmentation of amygdala subregions in children as young as six years of age (Kim et al., 2010, Qin et al., 2012). Given that we had no *a priori* lateralization hypotheses, we averaged signal from right and left amygdala masks. FC of amygdala subregions

was determined by semipartial correlation using the CONN Functional Connectivity Toolbox (ver.12.p; www.nitrc.org/projects/conn). Between group effects were considered within an anatomically defined medial prefrontal region used in prior works (Etkin and Schatzberg, 2011, Marusak et al., 2014b). This mask was selected to encompass pregenual (pgACC) and subgenual (sgACC) regions of the anterior cingulate, as these regions suppress limbic reactivity through direct connections to the amygdala. Family-wise error (FWE) corrected $p < 0.05$, significance level was used.

Motion poses a significant source of noise in FC analyses. None of the participants included in the present study had motion exceeding 1.5 mm in any direction. We addressed residual motion-related artifacts in three steps. First, Siemens MRI motion correction (MoCo) software was used during image acquisition. This procedure retrospectively measures 6 parameters of rigid-body translation and rotation, and produces a corrected time series using affine transformation. Second, functional image volumes were realigned to the mean image in SPM8. Third, realignment parameters (with another six parameters representing their first order temporal derivatives) were removed with covariate regression analysis before computing amygdala FC. Signals from white matter and cerebral spinal fluid were also regressed out using anatomical component correction (aCompCor; Behzadi et al., 2007, Chai et al., 2012). Low overall movement levels and lack of differences between groups augment confidence that motion did not compromise observed effects.

Secondary whole-brain analyses were performed to comprehensively evaluate connectivity of the amygdala. In addition to CM and BL regions, a superficial (SF) amygdala subregion mask was generated using procedures described above, and connectivity from this area was also examined for possible differences between groups. Regions showing altered FC

between groups were reported at a threshold of $p < 0.005$, cluster minimum = 10 voxels. This threshold was derived from suggested standards for whole-brain comparisons (Lieberman and Cunningham, 2009). Results that survived multiple comparisons correction for the whole brain (spatial cluster extent threshold; > 158 voxels for $p < 0.05$ FWE corrected) are denoted with an *asterisk in Table S1 (see the online data supplement[2]). To plot the direction of trauma-related amygdala connectivity, individual participant beta values were extracted from peaks using 4 mm radius spheres. Pearson correlation was used to test for associations between amygdala FC and anxiety, depressive symptoms, IQ, and income. All coordinates are reported in Montreal Neurological Institute (MNI) convention.

To further validate our findings, data were re-analyzed using motion "scrubbing" (Power et al., 2012). Specifically, movement was plotted and visually inspected for each participant using ArtRepair software (http://cibsr.stanford.edu/tools/human-brain-project/artrepair-software.html). Through this process we identified 4 trauma and 3 comparison participants that required censorship. High-movement frames were removed, and following this, maximal movement for all individuals in the sample was reduced to 0.171 mm and 0.392 degrees. Major comparisons and study outcomes were evaluated using this alternative analytic scheme, and those data are reported in the online data supplement.

3.3 Results

3.3.1 Participants

3.3.1.1 Demographics and movement. As shown in Table 2, groups did not differ in age, pubertal maturation, sex, race/ethnicity, parental educational attainment, or movement during the resting-state scan. Overall, residual movement was well within accepted standards (< 1.5 mm

[2]http://scan.oxfordjournals.org.proxy.lib.wayne.edu/cgi/pmidlookup?view=long&pmid=25836993

translational rms; cf. Fair et al., 2012). Relative to comparison youth, trauma-exposed participants reported lower levels of household income and IQ (see Table 2), effects that were anticipated based on prior work (e.g. De Bellis, 2001, Lantz et al., 2005). Follow-up analyses controlling for income and IQ were conducted on FC data, as described below. There were significantly more females than males represented across the entire sample, ($\chi^2_{(1)}$= 6.1, p = 0.014). Distribution of participant race/ethnicity across the sample was representative of the study community (Wayne Country, Michigan; $\chi^2_{(2)}$= 4.09, p = 0.13; www.census.gov). 4 participants (3 trauma, 1 comparison) were on psychotropic medications (1 on stimulants, 1 on selective serotonin reuptake inhibitors, 2 on serotonin-norepinephrine reuptake inhibitors, and 1 on beta-2 adrenergic agonists). Medications were not withheld for scanning. Follow-up analyses excluding participants on medications yielded no changes to observed effects.

Table 3. Demographic and Clinical Characteristics by Group

Variable	Trauma (n = 21)	Comparison (n = 21)	Group Comparison (p-value)
Age, m (SD)	12.77 (2.00)	12.32 (2.19)	ns
Pubertal Maturation (Tanner Stage), m (SD)	3.75 (0.95)	3.12 (1.38)	ns
Sex (Female), n (%)	15 (71.43)	14 (66.67)	ns
IQ, m (SD)	90.55 (10.9)	109.94 (16.04)	< 0.001
Race/Ethnicity, n (%)			
African American	10 (47.62)	10 (47.62)	ns
Caucasian	4 (19.05)	10 (47.62)	
Hispanic	3 (14.28)	0 (0)	
Not reported	4 (19.05)	1 (4.76)	
Annual Household Income, n (%)			
Less than $40,000	17 (81)	10 (47.6)	0.018
$40,000 - $60,000	1 (4.8)	7 (33.3)	
$60,000 - $80,000	2 (9.5)	1 (4.8)	
$80,000 - $100,000	0	1 (4.8)	
Over $100,000	0	2 (9.5)	

Not reported	1 (4.8)	0	
Highest Level of Parental Education, n (%)			
No GED/ no high school diploma	2 (9.5)	1 (4.8)	
GED/ high school diploma	4 (19)	1 (4.8)	
2-year degree or some college	8 (38.1)	9 (42.9)	
4-year degree	3 (14.3)	7 (33.3)	ns
Masters	3 (14.3)	2 (9.5)	
Doctorate	0	1 (4.8)	
Not reported	1 (4.8)	0	
Type of Trauma Endorsed[a], n (%)			
Physical abuse	4 (19%)	0	
Neglectful home environment	3 (14%)	0	
Emotional abuse	2 (10%)	0	
Exposure to domestic violence	14 (67%)	0	
Exposure to any other violence not already identified	11 (52%)	0	
Multiple separations from parent or caregiver	4 (19%)	0	
Sexual abuse or exposure	5 (24%)	0	
Anxiety Symptoms (SCR)[b], m (SD)	22.97 (16.49)	14.23 (11.08)	0.05
Depressive Symptoms (CDI)[c], m (SD)	4 (5.32)	1.8 (2)	ns
Motion During Scan			
Translational max frame-to-frame excursion (SD)	0.66 (0.31) mm	0.56 (0.25) mm	
Rotational max frame-to-frame excursion (SD)	33.8 (15.47)°	32.09 (18.91)°	
Translational mean movement (SD)	0.13 (0.08) mm	0.1 (0.6) mm	ns
Rotational mean movement (SD)	6.3 (3.44)°	6.87 (4.58)°	
Translational rms (SD)	0.1 (0.05) mm	0.08 (0.02) mm	
Rotational rms (SD)	< 0.05 (< 0.05)°	< 0.05 (< 0.05)°	

Note: [a]Trauma criteria are from the Children's Trauma Assessment (CTA) Center Screen Checklist (Item 1) by the Michigan Trauma Assessment Center. [b]SCR = Screen for Child Anxiety-Related Emotional Disorders. [c]CDI = Children's Depression Inventory. All p-values derived from t-tests with the exception of sex and race/ ethnicity comparisons, which used chi-square tests, and income and parental education, which used Mann-Whitney U tests. Abbreviations: m, mean; SD, standard deviation; n, number; ns, not significant; max, maximum; rms, root mean squared head position change during the resting-state scan.

3.3.1.2 Self-reported affect measures. Although the trauma group was not chosen on the basis of psychopathology, participants with histories of trauma reported higher levels of anxiety (SCR) relative to comparison participants (Table 3). This is consistent with reports that childhood trauma exposure is a strong predictor of emotional psychopathology (Kessler et al., 1997). Follow-up analyses were conducted to account for effects of anxiety on connectivity (see below). While anxiety scores reported during lab and MRI visits were significantly correlated within subject (see *Methods*), it is possible that variation across visits differed between groups. We therefore tested for effects of visit on anxiety levels using a Group (trauma, comparison) x Visit (lab, MRI) ANOVA. Consistent with group differences reported above, a significant main effect of Group emerged, $F(1, 80) = 7.08$, $p < 0.009$. No main effect of Visit, or Group x Visit interaction was observed (p's > 0.4) suggesting that trait anxiety was stable over a period of two weeks and variability between visits did not differ between groups. In contrast, average state levels of fear/anxiety during the MRI visit (VAS) did not significantly differ between groups, $t(39) = 0.3$, $p = 0.77$. Given that VAS ratings were previously shown to correlate with cortisol reactivity during the scan (Thomason et al., 2013), this result suggests that connectivity differences are not likely influenced by group differences in biologic stress responsivity during the MRI visit. Groups did not differ on trait levels of depressive symptoms, $p > 0.08$ (see Table 3).

3.3.2 Lack of amygdala-sgACC connectivity in trauma-exposed youth

We observed significant group differences in CM amygdala-sgACC connectivity ($x = 8$, $y = 18$, $z = -8$, pFWE = 0.022, $Z = 3.80$). Extraction of average connectivity strength within this cluster revealed that the group effect resulted from the predicted negative amygdala-sgACC FC in comparison participants, which was absent in trauma-exposed youth (Figure 4). CM

amygdala-sgACC FC was not related to anxiety (SCR, $r(42)= 0.01$, $p = 0.92$) or depressive (CDI, $r(42) = 0.03$, $p = 0.83$) symptoms across the sample, or within the trauma group (SCR, $r(21) = -0.22$, $p = 0.34$; CDI, $r(21) = -0.15$, $p = 0.51$). Group differences in CM amygdala-sgACC connectivity remained significant when controlling for IQ (peak at $x = -12$, $y = 40$, $z = -4$, pFWE = 0.025, $Z = 3.77$) and income (peak at $x = 10$, $y = 22$, $z = -10$, pFWE = 0.027, $Z = 3.74$). No significant effects of trauma were observed for BL amygdala-sgACC connectivity at $p < 0.05$ FWE-corrected.

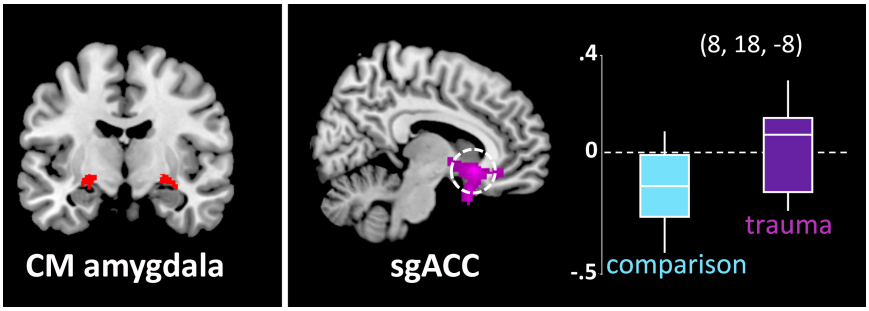

Figure 4. Absent negative centromedial (CM) amygdala-subgenual cingulate (sgACC) negative connectivity in trauma-exposed youth. Left: Signal was averaged across anatomically-defined bilateral CM amygdala source region. Right: Tukey's boxplots depict connectivity values by group centered on the sgACC peak. The middle line indicates the median, vertical line the range, and the limits of the box represent upper and lower quartiles. Results significant at a threshold of $p < 0.05$, FWE-corrected.

3.3.3 Divergent patterns of amygdala subregion FC across the sample

Secondary whole-brain analyses were performed to examine connectivity of major amygdala subregions. FC maps across the entire sample (Figure 5) revealed unique patterns of connectivity for bilateral CM, BL, and SF amygdala subregions that are consistent with previous cytoarchitectonically-based amygdala FC studies (e.g. Roy et al., 2009, Brown et al., 2014). Briefly, CM showed signal covariation with striatal regions, whereas BL showed signal

covariation with temporal and frontal cortical regions. SF signal was strongly correlated with signal in limbic regions.

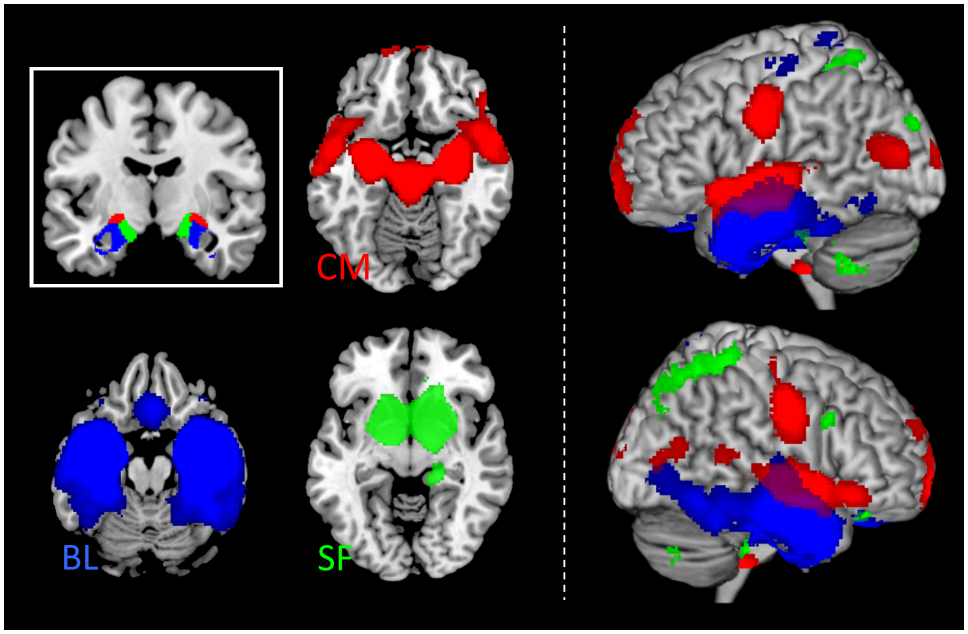

Figure 5. Functional connectivity of amygdala subregions across the entire youth sample. Left: Inset shows anatomically-defined bilateral amygdala seed regions: CM (centromedial; red), BL (basolateral; blue), and SF (superficial; green). Axial slices show brain areas positively correlated with amygdala seed regions. Right: Surface renderings are used to depict separation and overlap of amygdala neural networks. Connectivity maps displayed at $p < 0.005$, cluster minimum = 10 voxels.

3.3.4 Trauma effects on amygdala whole-brain connectivity

Extensive differences in amygdala FC were observed between groups (see Table S1 in online data supplement). We also found that in a subset of areas in which groups differed, FC also related to income, IQ, anxiety, or depressive symptoms. Thus, not only does amygdala FC vary across brain areas, but this variation relates to symptom severity. Data are summarized in Figure 6 as radar plots, where average FC is plotted for each group, by region. Visual inspection of these plots suggests that, overall, individuals who have experienced trauma demonstrate increased positive connectivity across widespread brain regions. Consistency was observed

across amygdala subregions in that frontoamygdala connectivity was negative in the comparison group but not in trauma-exposed youth (see Figure 6). This was also true in anterior insula-amygdala connectivity, that was more negative in comparison but not in trauma-exposed participants for the SF subregion (Figure 7). A different effect was observed for the dACC; there, we observed increased positive amygdala connectivity in comparison but not trauma participants (Figure 7).

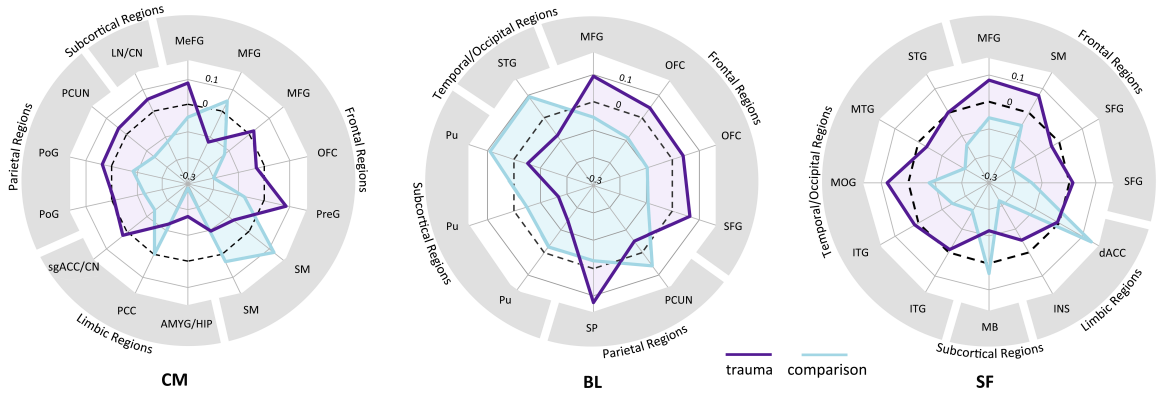

Figure 6. Differential patterns of amgydala subregion functional connectivity (FC) across the whole brain in trauma-exposed (purple) and comparison (blue) youth. Numerical values represent the average within-group correlation strength in areas showing significant group differences at the whole brain level, across frontal, limbic, parietal, subcortical, and temporal/occipital regions (coordinates are provided in Table S1, see online data supplement). Wider area on radial plots in trauma-exposed youth suggests a pattern of enhanced positive FC of the amygdala to many brain regions, particularly to areas of the frontal cortex. Dashed line indicates zero; the middle of the plot indicates negative values. Abbreviated brain regions are defined in Table S1 (see online data supplement).

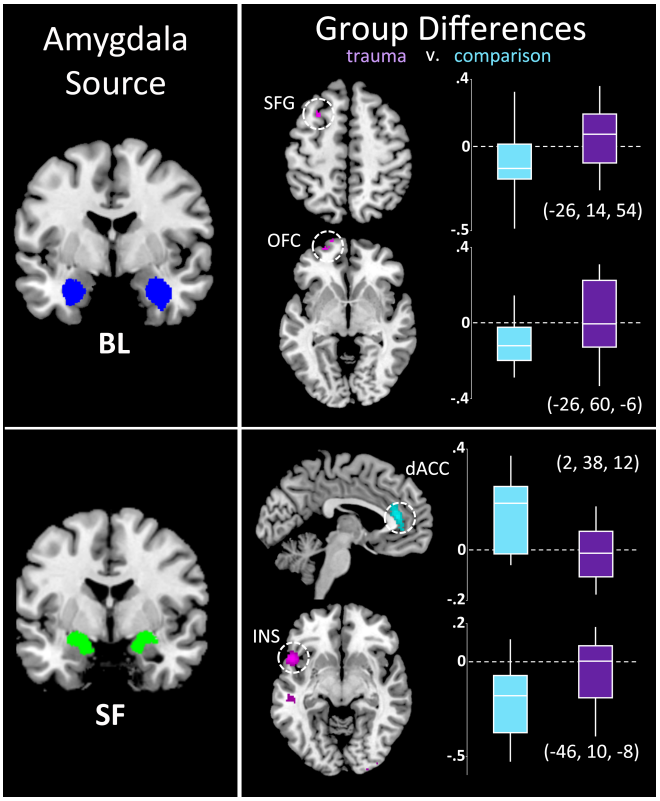

Figure 7. Regions showing group differences (trauma, purple; comparison, blue) in functional connectivity with BL (basolateral) and SF (superficial) amygdala. Right: Tukey's boxplots depict connectivity values by group. The middle line indicates the median, vertical line the range, and the limits of the box represent upper and lower quartiles. Corresponding MNI coordinates of peak group difference (see Table S1 in online data supplement) are indicated adjacent to each boxplot. Results are shown at $p < 0.005$, cluster minimum = 10 voxels, for display purposes. Abbreviations: SFG, superior frontal gyrus; OFC, orbitofrontal cortex; dACC, dorsal anterior cingulate; INS, insula.

3.3.5 Replication of results in scrubbed data

Data were reanalyzed utilizing censoring of high-movement frames. Following censoring, average group motion was < 0.05 mm and < 0.002 degrees rms. As shown in Figure S1 (see online data supplement), main findings of amygdala FC across the sample (Figure 5), and group differences in FC in the sgACC (Figure 4), superior frontal gyrus, dACC, and insula (Figure 7) were replicated. However, the group difference in BL amygdala-orbitofrontal cortex connectivity was no longer significant.

3.4 Discussion

The present study evaluates links between early life trauma and neural circuit organization, within a high sociodemographic risk youth sample. Our results indicate reduced negative amygdala-sgACC FC in trauma-exposed youth, fitting with prior observed associations between disrupted amygdala circuitry and emotional psychopathology. For example, alterations in frontoamygdala circuitry have been reported in adults with PTSD (Brown et al., 2014), and adolescents (Roy et al., 2013) and adults with generalized anxiety disorder (Etkin et al., 2009). We add to this conceptualization by discovering that experience (trauma) is associated with altered intrinsic amygdala FC in formative, developmental years. In particular, trauma-related changes were observed within amygdala-sgACC circuitry, a pathway that is critical for emotion regulation. These findings are in line with our recent study showing a reduced ability to regulate emotional conflict and an absence of negative frontoamygdala connectivity during conflict resolution in trauma-exposed youth (Marusak et al., 2014b). The present findings extend this work by demonstrating that deficits in this critical emotion regulatory pathway are present even when individuals are not engaging regulatory control processes.

Four studies most similar to the current investigation are summarized in Table S2 (see the online data supplement). The most comparable study, implemented by Gee and colleagues, evaluated FC in orphanage-reared youth during a face processing task (Gee et al., 2013a). Focusing on the same frontoamygdala circuitry emphasized here, they too found abnormalities in neural FC between groups. Interestingly, their effects were related to the type of emotional face being processed. That is, between group differences were noted during the processing of fear faces, but the direction of effects differed during processing of happy faces. This result, paired with data presented here about intrinsic FC during resting-state suggests that dynamics in

frontoamygdala circuits in trauma-exposed youth may relate to processing demands, and results obtained should be considered in that light. Gee and colleagues also provide evidence that the age of participants being examined may interact with observed effects. They found that comparison and orphanage-reared participants exhibited different patterns of age-related change in frontoamygdala FC to fear faces. Specifically, between group differences appear to have been significant in children but not adolescents. This could be interpreted as a reduction in differences between groups with age. Though the cross-sectional nature of this sample precludes direct evaluation of this possibility, this observation can be regarded in consideration of current theory that principals of neural connectivity at one point in life may be adaptive, while at another may confer risk (Tottenham, 2013).

We interrogated amygdala FC from three amygdala subregions. CM amygdala is involved in allocating attention to relevant stimuli (Davis and Whalen, 2001) and mediating increased vigilance (Dringenberg and Vanderwolf, 1996). In contrast, BL amygdala has been linked to associative learning processes, and the SF amygdala has been implicated in social/affective processing (LeDoux, 2003, Phelps and LeDoux, 2005, Goossens et al., 2009). Given that amygdala subregions exhibit unique developmental patterns of FC across childhood and adolescence (Gabard-Durnam et al., 2014), it is likely that trauma impacts these circuits in different ways. We support this supposition by having attained commonalities and discrepancies in effects across subregions. Commonalities were observed across amygdala subregions such that youth that endured traumatic experiences tended to have diminished frontoamygdala connectivity. Discrepancies were noted for regions of the striatum, parietal regions, and regions comprising the default mode brain network (e.g., precuneus and posterior cingulate). For these areas, CM and BL showed trauma-related FC effects but SF did not. In contrast, analysis of SF

connectivity revealed trauma effects in regions that comprise the salience network of the brain, the insula and dACC, whereas other subregions did not. Thus, amygdala subregions critical for learning and attention and for social/affective processing demonstrate overlapping, but also distinct trauma-related alterations in connectivity.

Our results support a model of reduced connectivity in emotion regulatory circuitry in urban youth with histories of trauma. Amygdala-medial prefrontal correlations are negative at rest (Gee et al., 2013b) and during the regulation of emotional responding (Hare et al., 2008), a pattern of connectivity thought to index top-down regulatory control. Behaviorally, loss of inhibitory affective control has been associated with exposure to trauma (Pechtel and Pizzagalli, 2011) as well as presence of clinical mood disorders (Etkin et al., 2013). Prior research has also described negative attention bias and augmented vigilance in those with histories of early life trauma exposure (Pollak, 2008). Furthermore, studies using task-based fMRI show hyperactivity of the amygdala and/or hypoactivity of medial prefrontal regions in individuals with anxiety (McClure et al., 2007), PTSD (Etkin and Wager, 2007), and in children with histories of early adversity (Tottenham et al., 2011, McCrory et al., 2013), which may reflect failure of prefrontal regions to regulate amygdala reactivity. The direction of our effects (more negative frontoamygdala FC in comparison participants) suggests reduced top-down control via frontoamygdala neurocircuitry may be a consequence of early traumatic experiences.

Whole-brain exploratory analyses identified stronger amygdala-anterior insula connectivity in trauma-exposed youth. Increased FC indicates greater signal covariance between regions that detect threat and generate fear responses (i.e., amygdala; LeDoux, 2003) and process meaning and prediction of aversive bodily states (i.e., insula; Craig, 2011). Stronger amygdala-insula FC has also been reported in adults with PTSD (Rabinak et al., 2011) and adolescents with

GAD (Roy et al., 2013), highlighting the relevance of this pathway for clinical disorders. Moreover, task-based fMRI studies have demonstrated increased amygdala and insula reactivity in children exposed to violence (McCrory et al., 2011) and soldiers that endure combat stress (van Wingen et al., 2011). Increased functional covariance in the amygdala and insula at rest suggests they may be primed for rapid co-activation, or that functions subserved by these regions tend to be more coordinated.

Our results indicate significantly reduced (positive) amygdala-dACC connectivity in trauma-exposed youth. Whereas ventral aspects of the ACC (e.g., pregenual and subgenual regions) are implicated in emotion regulation, dACC is associated with the expression of fear (Etkin et al., 2011). Evidence suggests that top-down (i.e., dACC/lateral prefrontal-based) forms of emotion regulation work by recruiting the ventral vACC, which dampens limbic reactivity directly (Etkin et al., 2011). Fitting with this conceptualization, our data show congruent between group effects in dACC and sgACC. Dorsal aspects of the ACC are also implicated in sympathetic nervous arousal (Critchley, 2005). Thus, our altered amygdala-dACC FC results may reflect aberrant modulation of autonomic nervous system function. This is consistent with reports of increased arousal and fear in trauma-exposed youth, which may contribute to the development of clinical disorders (e.g., anxiety; Glaser, 2000).

We observed that differences in neuroconnectivity between trauma-exposed and control groups were associated with depression or anxiety symptomology only in select regions, including the sensorimotor cortex, middle temporal gyrus, and superior parietal cortex. This is highly consistent with the observation that trauma exposure does not precisely predict emergence of psychopathology. Considerable data has shown that outcomes resulting from trauma exposure vary greatly across individuals (Cicchetti and Rogosch, Felitti et al., 1998, Tottenham and

Sheridan, 2009). Thus, one would not expect that the relationship between brain measures and previously experienced trauma would be ubiquitously significantly correlated. These variables are neither orthogonal, nor perfectly correlated. Thus, having observed some but not complete overlap in significance could have been predicted. Similar consideration could be predicted for observed associations in income and IQ, all of which are presented in superscript in Table S1 (see the online data supplement).

Given increased awareness that motion confounds interpretation of fMRI data and the resulting shift toward rigorous approaches to dealing with motion in resting state data, we reanalyzed all data using an alternative scrubbing approach (Deen and Pelphrey, 2012, Van Dijk et al., 2012, Power et al., 2015). We saw that observed main effects were consistent, with the exception of a group difference in the orbitofrontal cortex that did not hold after scrubbing. It is possible this result did not replicate because the group difference covered a smaller territory to begin with (15 voxels), and because clusters smaller than 10 contiguous voxels are not reported as significant. The reason that in large part we saw consistent effects in scrubbed and non-scrubbed data may be due to the low overall movement profile of participants in the original analyses.

Our results underscore that traumatic stress may alter corticolimbic circuitry in the immature brain and this may precede the onset of clinically significant conditions. There is evidence suggesting that changes in frontoamygdala circuits persist even decades later into adulthood (Dannlowski et al., 2012), emphasizing the relevance of early life exposures for lifelong socioemotional functioning. Pharmacological treatment (i.e., fluoxetine) has been shown to normalize alterations in frontamygdala connectivity in depressed adults (Chen et al., 2008), an encouraging result. While altered amygdala connectivity may reflect ontogenetic adaptation to an

unsafe rearing environment, it is possible that these neural changes confer elevated threat vigilance, which may be detrimental to healthy emotional development. For instance, it has been suggested that increased amygdala-insula FC mediates anxious anticipation of negative events (Carlson et al., 2011). Recalibration of neural connectivity in corticolimbic circuitry in early life may be detrimental to evaluation of threat and safety, and compromise a child's ability to master age-appropriate skills in social and cognitive domains. For example, altered connectivity may reflect changes in synaptic function within this emotion regulatory network or reduced priming of network components that allow for rapid control of emotional responding.

Our findings should be considered in light of limitations. First, we were not sufficiently powered to differentiate results based on onset (age) or type of trauma. While retrospective analysis shows that trauma onset and trauma type relate to distinctive patterns of emotional functioning (English et al., 2005), prior studies also document nonspecific effects of trauma type on outcomes (Arata et al., 2007, Collishaw et al., 2007) and some suggest that disentangling unique effects may result in overly narrow interpretations (Green et al., 2010). Next, participants were drawn from a larger study, and were not selected on the basis of IQ or income. As a result, groups were not matched on these variables. This is not surprising given the strong association between these variables and trauma prevalence (De Bellis, 2001), but also not ideal for disentangling connectivity effects. However, the alternative is to select matched samples that may not convey the natural conditions present in trauma with regard to neural connectivity. Moreover, we are assured by the consistency of our findings with both animal and human research (Gee et al., 2013a, Malter Cohen et al., 2013a, Brown et al., 2014), and because follow-up analyses indicated that amygdala-sgACC results held when controlling for income and IQ. In addition, we provide areas that are influenced by trauma exposure and other risk factors (i.e.,

poverty, IQ, anxiety, depression). Interactions between these variables are areas for future research. While we did not find an association between frontoamygdala connectivity and anxiety or depressive symptoms, it is possible that additional symptom-brain associations would be detected with a larger sample. Along this line, an additional consideration is the sample size is relatively small (n = 42), and findings are correlational in nature - thus precluding ability to make causal attributions about changes in frontoamygdala connectivity and the experience of childhood trauma. Future longitudinal examinations of amygdala connectivity in larger samples will be necessary to better understand mechanisms contributing to neural circuit reorganization following early life stress. Finally, poor spatial resolution of standard fMRI acquisitions and susceptibility of the amygdala to EPI image distortions and draining vein effects (Merboldt et al., 2001) may lead to spatial localization errors. Furthermore, use of an 8 mm smoothing kernel to investigate small volumes such as amygdala subregions is not an optimal design. Despite these considerations, cytoarchitechtonically-based amygdala FC analyses have yielded consistent, replicated delineation of differential connectivity of major amygdala subregions (Etkin et al., 2009, Roy et al., 2009, Roy et al., 2013, Brown et al., 2014). Also, the fact that patterns observed here replicate prior neuroimaging and anatomical work in animals affords further confidence in the approach.

3.4.1 Conclusions

Reduced negative amygdala-sgACC connectivity was observed in a sociodemographic risk sample of youth exposed to severe trauma. Isolation of these effects augments prior studies in adults obtaining similar results by evaluating intrinsic connectivity in formative years (e.g., childhood) and by measuring change in a sample at substantially increased risk for developing clinical syndromes (i.e., urban, low income, minority). Our results support the supposition that

the biological embedding of adversity in early life may include changes in neural connectivity, which in turn may alter interactions with the world and susceptibility to disease.

CHAPTER 4: CHILDHOOD TRAUMA DISRUPTS THE AUTOMATIC REGULATION OF EMOTION PROCESSING

4.1 Introduction

Converging evidence suggests that trauma exposure, particularly in early life, fundamentally alters the way emotional information is processed and prioritized. Indeed, behavioral studies in children with histories of trauma show enhanced attention to and difficulty disengaging from emotional stimuli (Tottenham et al., 2010). Early life trauma exposure is a potent risk factor for neuropsychiatric disorders including anxiety, depression, and posttraumatic stress disorder (Kaufman et al., 2000, Gilbert et al., 2009), which are also hallmarked by abnormalities in the processing and regulation of emotion (Etkin and Wager, 2007). Examining emotion regulation in trauma-exposed children and adolescents (youth) who are at elevated risk for developing emotional psychopathology may illuminate biological pathways that link trauma exposure in early life to subsequent emergence of clinical disorder.

Behavioral studies in trauma-exposed individuals report deficits in cognitive control and emotion regulation (Tottenham et al., 2010, Pechtel and Pizzagalli, 2011), processes that are known to develop across the first two decades of life (Casey et al., 1997). Brain regions subserving these functions (e.g., pgACC, dorsolateral prefrontal cortex [DLPFC]) also show protracted development, and are particularly vulnerable to the deleterious effects of early life stress (Pechtel and Pizzagalli, 2011). It is therefore critical to identify the neural and behavioral correlates of trauma exposure during a time when brain circuits are still forming, and when interventions can be maximally impactful. Yet, neurobiological research examining emotion regulatory pathways in youth exposed to trauma is limited.

Emerging research supports the notion that behavioral alterations observed in trauma-exposed youth may relate to changes in underlying neurobiological processes. For instance,

children with early life trauma exposure show exaggerated amygdala response to threatening cues (McCrory et al., 2011), and altered connectivity in brain systems relevant for detecting and processing emotional information (Gee et al., 2013a, Thomason et al., In Press). Together, it appears that early life adversity is associated with reorganization of neural circuits in ways that enhance processing of salient emotional stimuli. Altered processing of emotional information may underlie or potentiate emotion regulation deficits reported in individuals with histories of early life trauma (see Pollak, 2008 for a review).

One way to examine emotion regulatory control processes is through neuropsychological tasks where emotional distracters are in direct conflict with task-relevant emotional information (see Fig.1). Emotionally incongruent stimuli, such as the word 'FEAR' superimposed on a happy face, cause reduced accuracy and greater reaction time (RT) interference (i.e., slowdowns). The amount of emotional 'conflict', or interference generated by incongruent stimuli (incongruent minus congruent trials [I-C]), is associated with activity in the amygdala (Etkin et al., 2006), a brain region that monitors and detects salient environmental stimuli (Whalen, 1998), and DLPFC, which may reflect effortful attentional control (Derrfuss et al., 2005).

When individuals engage emotion regulatory systems, they become more prepared to inhibit immediate subsequent conflict. As a result, accuracy is improved and RT interference is reduced for incongruent trials if they are preceded by another incongruent trial rather than a congruent trial (iI-cI; Botvinick et al., 1999, Botvinick et al., 2001). This effect, termed "conflict regulation", seems to occur outside of conscious awareness and thus is a type of automatic emotion regulation (Etkin et al., 2010). Prior fMRI and lesion studies show that automatic emotional conflict regulation relies on inhibited processing of emotional distracters through top-down (i.e., pgACC) modulatory control of amygdala responsivity (Etkin et al., 2006, Egner et al.,

2008, Maier and di Pellegrino, 2012). Perturbations in this critical emotion regulatory pathway have been observed in adults with anxiety and depression (Etkin and Schatzberg, 2011), and may underlie emotion regulatory deficits observed in adults with early life trauma (Ford, 2005).

The present study examines the spontaneous regulation of emotional processing in a sample of trauma-exposed, urban, low-income, minority youth at high risk for psychopathology. Here, we define childhood trauma as the experience of event(s) that threaten a child's safety (i.e., witness violence), undermine their security (i.e., neglect), or fragment attachment bonds (i.e., abuse). We utilized an adapted version of the emotional conflict task (Etkin et al., 2006) for children, by using salient peer emotion faces. Based on behavioral research in trauma-exposed youth (Tottenham et al., 2010) and prior applications of the emotional conflict task in adults with anxiety and major depressive disorder (Etkin et al., 2010, Etkin and Schatzberg, 2011), we hypothesized that trauma-exposed youth would exhibit greater deficits in ability to ignore emotional distracters that are task irrelevant, and reduced ability to regulate emotional conflict. Our analyses focused on amygdala-pgACC circuitry, given prior work emphasizing that this pathway underlies emotional conflict regulation through inhibition of amygdala reactivity (Etkin et al., 2006). Specifically, we predicted higher amygdala response to conflict, lower pgACC response during conflict regulation, and lower negative amygdala-pgACC functional connectivity during conflict regulation in trauma-exposed youth. We also tested for the presence of compensatory regional responses in the DLPFC during conflict regulation - previously observed in depressed but not anxious adults (Etkin and Schatzberg, 2011). Finally, we tested whether neural function mediates the association between trauma exposure and emotional well-being. In particular, we evaluated anxiety and depression symptoms, and trait RS. The latter was of interest because of emerging research showing that diminished RS is a promising marker of

vulnerability to affective disorders following stress (Bogdan et al., 2013). Reduced emotion regulatory ability may be associated with changes in reward function. We therefore tested if higher amygdalar responses to conflict mediated the association between trauma exposure and low RS.

4.2 Methods

4.2.1 Participants

A total of 51 children and adolescents, recruited through online advertisements or child psychiatry clinics (Detroit, Michigan), participated in this fMRI study. Exclusionary criteria included: English as a second language, lower than a 2nd grade reading level, history of brain injury, neurological or movement disorders, or presence of MRI contraindications. Parental informed written consent and child/adolescent assent were obtained prior to participation.

4.2.2 Trauma and Clinical Measures

Using both parent and child report, participants who endorsed (lifetime) at least one trauma itemized on the Children's Trauma Assessment Center Screen Checklist (source: Michigan Trauma Assessment Center) were categorized as 'trauma'. Number and type of endorsed traumas are provided in Table 4. Participants with movement > 4 mm in the scanner (n = 13; 3 trauma, 10 comparison), accuracy < 50% (n = 5; 1 trauma, 4 comparison), or errors in behavioral data collection (n = 2; 1 trauma, 1 comparison), were excluded from analyses. Therefore, all neuroimaging data are reported for 14 trauma-exposed and 16 age-, sex-, and IQ-matched comparison youth. For completeness, all participants tested with usable behavioral data were included in behavioral analyses. This resulted in the addition of two participants (1 trauma, 1 comparison) that did not qualify for neuroimaging analyses (n = 15 trauma; n = 17 comparison). IQ was assessed using the Kaufman Brief Intelligence Test (KBIT v.2; Kaufman

and Kaufman, 2004a). Participant ages (9-16) were selected to align with the emergence of puberty; puberty has been identified as a time when neuropsychiatric disorders frequently manifest (Angold et al., 1998). Pubertal development was assessed using Tanner staging. Following prior work (Forbes et al., 2009), participants were categorized as pre/early (Tanner stages 1-2) or mid/late pubertal (stages 3-5). Effects of pubertal maturation on trauma-related group differences were examined.

Table 4. Participant demographics by group

	Trauma (n = 14)	**Comparison (n = 16)**
Age, m (SD)	12.7 (2.09)	12.76 (2.21)
Sex (female), n (%)	10 (71.4)	14 (87.5)
IQ, m (SD)	100 (13.27)	102.57 (13.8)
Pubertal Maturation, n (%)		
Pre/early pubertal (Tanner stages 1-2)	5 (35.7)	5 (31.2)
Mid/late pubertal (Tanner stages 3-5)	9 (64.3)	11 (68.8)
Race/Ethnicity, n (%)		
African American	5 (35.71)	9 (56.25)
Caucasian	3 (21.43)	5 (31.25)
Hispanic	2 (14.29)	0
Biracial	1 (7.14)	1 (6.25)
Not reported	3 (21.43)	1 (6.25)
Household Annual Income, n (%)		
Less than $40,000	11 (78.57)	7 (43.75)
$40-60,000	1 (7.14)	5 (31.25)
$60-80,000	1 (7.14)	2 (12.5)
Over $80,000	0	2 (12.5)
Not reported	1 (7.14)	0
Type of Trauma Endorsed, n (%)		
Physical abuse	2 (14)	0
Neglectful home environment	3 (21)	0
Exposure to domestic violence	7 (50)	0
Exposure to any other violence not already identified	7 (50)	0
Multiple separations from parent or caregiver	2 (14)	0
Sexual abuse or exposure	3 (21)	0
Anxiety Symptomology (SCR), m (SD)	19.29 (13.67)	15.18 (11.38)
Depressive Symptomology (CDI), m (SD)	2 (2.57)	2.12 (2.47)
Reward Sensitivity (normalized; BAS), m (SD)	0.06 (0.56)	-0.25 (.84)
Reward responsivity	17.47 (1.34)*	19.73 (1.58)*

Fun seeking	12.21 (1.89)	12.53 (2.85)
Drive	10.85 (2.54)	11.33 (2.06)
Motion During Scan[a], m (SD)		
Translational mean movement	0.07 (0.06)	0.04 (0.02)
Rotational mean movement	5.7 (5.7)	2.86 (2.86)
Translational RMS	0.04 (.02)	0.04 (.02)
Rotational RMS	0.05 (.03)	0.04 (.03)

**Indicates group comparison is significant at $p \leq .05$. Chi-square tests were used for sex, race/ethnicity, income, and trauma-type comparisons; two-sample t-tests for age, psychopathology, and motion comparisons.*
[a]Translational (x, y, z) movement is reported in mm; rotational, in degrees. Abbreviations: mean, m; Intelligence Quotient, IQ; Screen for Child Anxiety Related Emotional Disorders, SCR; Children's Depression Inventory, CDI; Behavioral Activation Subscale of the BIS/BAS scales, BAS; root-mean-square head movement, RMS.

Three validated self-report measures of symptoms and affective traits were administered: (1) the 41-item Screen for Child Anxiety-Related Emotional Disorders (SCR; Birmaher et al., 1997), (2) the 10-item Children's Depression Inventory (CDI; Kovacs, 1985), and (3) the Behavioral Inhibition and Activation Scales (BIS/BAS; Carver and White, 1994). Following prior work (Garner et al., 2012), RS was conceptualized as the BAS component of the BIS/BAS. The BAS is comprised of three subscales: drive, fun seeking, and reward responsiveness. These subscales are intercorrelated (*r's* > 0.3, *p's* ≤ 0.05 in the present sample), but relate to discrete aspects of reward behavior. BAS_{drive} captures the strength with which reward outcome guides subsequent behavior, and has been used as a measure of trait reward-seeking (Hickey et al., 2010). $BAS_{fun\ seeking}$ is akin to trait novelty-seeking, whereas reward responsiveness (BAS_{rr}) indexes the degree to which a person derives pleasure from reward. We tested each of the BAS subdimensions, as well as a combination of the three (Garner et al., 2012) using *z*-scores to form an overall index of RS. A visual analog scale (VAS) was used to obtain an average rating of fear/anxiety during the MRI visit (repeat measures at 30-minute intervals) as previously described (Thomason et al., 2013).

4.2.3 Experimental Paradigm

The task consisted of 163 presentations of happy or fearful facial expression photographs, overlaid with the words "FEAR" or "HAPPY" to create emotionally congruent and incongruent stimuli (see Figure 8). Participants were instructed to indicate the facial affect with a button press response, while trying to ignore the task-irrelevant word stimuli. Stimuli were presented for 1,000 ms, with a varying interstimulus interval of 2,000-4,000 ms (mean = 3,000 ms), in a pseudorandom order, counterbalanced across trial types for expression, word, response button, and gender. Participants were given a practice task before entering the scanner. Stimuli were presented with EPrime software v.2.0 (Psychology Software Tools, Inc., Pittsburgh, PA) during fMRI scanning and displayed on a back-projection screen that was viewed by the participants via a mirror attached to the head coil. Task duration was 12:46.

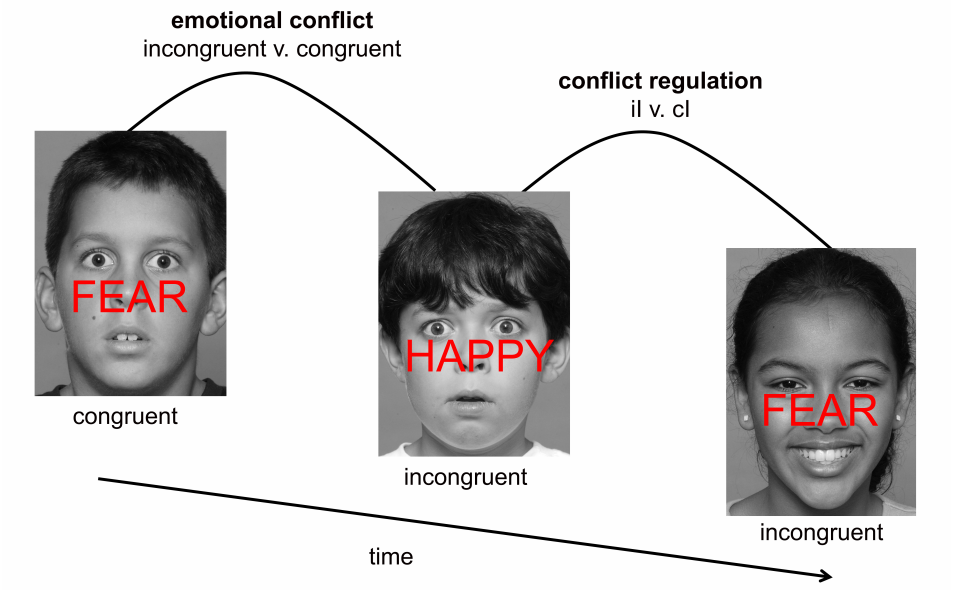

Figure 8. Emotional conflict task. Participants were instructed to identify the underlying facial emotion (fearful or happy) while ignoring an overlying emotion word ('FEAR' or 'HAPPY'). Trials varied such that emotional distracter words either matched ["congruent", (C)] or conflicted ["incongruent", (I)] with the underlying facial expression. Conflict interference was assessed by contrasting incongruent trials with congruent trials. Conflict regulation was isolated by

contrasting postincongruent incongruent (iI) with postcongruent incongruent (cI) trials. The task was adapted for children by utilizing an established set of child emotion-face stimuli of varied ethnicities, ages 10-17 (Egger et al., 2011). Importantly, the stimuli used matched the demographics of our study sample and minimized complex relations inherent in adult face stimuli (Marusak et al., 2013).

4.2.4 Behavioral Analysis

Accuracy and RT (correct trials) were analyzed in IBM SPSS v.22. Behavioral effects were considered significant at a $p \leq 0.05$ (two-tailed) threshold.

4.2.5 Imaging Data Acquisition

Functional images were acquired using a 3 Tesla Siemens Verio scanner equipped with a 12-channel head coil (MRI Research Center, Wayne State University). Twenty-nine axial slices were acquired across the whole brain using T2*-weighted echo-planar imaging (TR: 2000 ms, TE: 25 ms, matrix: 220 × 220, flip angle: 90°, voxel size: 3.44 × 3.44 × 4 mm). High-resolution anatomical images were acquired for individuals using a T1-weighted 3D MP-RAGE sequence (TR: 1680 ms, TE: 3.51 ms, orientation: axial, matrix: 384 × 384, 176 slices, flip angle: 9°, voxel size: 0.7 × 0.7 × 1.3 mm).

4.2.6 Movement during the Scan

During acquisition, Siemens MRI motion correction (MoCo) software was used to retrospectively measure 6 parameters of rigid-body translation and rotation for each time frame and produce a corrected time series using affine transformation. Following exclusion of high movement participants, movement fell within accepted standards (e.g., < 1.5 mm rms; Fair et al., 2012; see Table 4).

4.2.7 Imaging Data Analysis

fMRI data were preprocessed using SPM8 software (Statistical Parametric Mapping; http://www.fil.ion.ucl.ac.uk/spm/) implemented in MATLAB (MathWorks, Inc., Natick, MA),

following procedures described in our prior work (Etkin et al., 2006, Marusak et al., 2013). The first four image volumes were excluded from analysis to allow for signal equilibration effects. Preprocessing steps included: (i) image realignment, (ii) spatial transformation to the Montreal Neurological Institute (MNI) template using the participant-specific transformation parameters created by fitting mean functional images to the single reference EPI standard template (in SPM). Data were not resampled during normalization, thus retained the native resolution (3.44 x 3.44 x 4 mm) for subsequent analysis. (iii) Images were then spatially smoothed with a Gaussian kernel of 6 mm full width at half maximum.

A 128-second temporal high-pass filter was applied to the data, and temporal autocorrelation was estimated using a first-order autoregressive model. Two independent participant-level models were created in the context of a general linear model to examine effects of (1) conflict and (2) conflict regulation. In the first model, separate regressors for the stimulus events (convolved with a canonical hemodynamic response function) were created for incongruent (I) and congruent (C) trials. 82 experimental trials were incongruent, and 81 were congruent. For the second model, trial types were broken down based on the preceding trial type: regressors were created for postcongruent incongruent trials (cI), postincongruent incongruent trials (iI), postcongruent congruent trials (cC), and postincongruent congruent trials (iC). There were 38 cI trials, 44 cC trials, 37 iC trials, and 44 iI trials. All participant-level models included regressors of no interest corresponding to the six motion parameters, and modeled error and posterror trials separately. Participant-level contrasts isolated (1) conflict-related neural activity by comparing I-C trials, and (2) conflict regulation by comparing iI-cI trials.

Group-level random-effects two-sample *t*-tests were used to test for group differences in neural activity during (1) emotional conflict (incongruent minus congruent trials [I-C]), and (2)

emotional conflict regulation (post-incongruent incongruent minus post-congruent incongruent trials [iI-cI]). The contrast iI-cI isolates activity during conflict trials for which behavior differs by virtue only of expectation created by the previous trial type (i.e., previous trial is either congruent or incongruent).

A psychophysiologic interaction analysis (Friston et al., 1997) was conducted to test for group differences in amygdala-pgACC FC during conflict regulation. We used a bilateral amygdala seed, given that group differences in amygdala activation were observed in both hemispheres and because we did not have *a priori* predictions about laterality. First, the deconvolved time course was extracted from bilateral amygdala using a mask defined by FSL FIRST subcortical segmentation tool (Patenaude et al., 2011). Then, activity within the amygdala was regressed on a voxel-wise basis against the psychological variable of interest (i.e., the interaction term), with the physiological (i.e., amygdala time course) and psychological (i.e., iI > cI contrast) variables serving as regressors of non-interest. Results were submitted to a random-effects group analysis using two-sample *t*-tests.

Neural results are reported for *a priori* regions of interest, using small-volume correction, $p < 0.05$, family-wise error-corrected; FWE. Areas examined include: (i) left and right DLPFC 10 mm spheres (x = -47, y = 21, z = 29; x = 51, y = 21, z = 32); and (ii) pgACC, utilizing the anatomically-defined mask described in our prior work (Etkin and Schatzberg, 2011). All coordinates are reported in Montreal Neurological Institute (MNI) convention. For regions showing group differences, signal was extracted from 4 mm radius spheres (centered on the peak) to plot the effects of trauma and to test for associations with behavioral and clinical measures. Given that prior research has found trauma-related effects in bilateral amygdala (McCrory et al., 2011), left and right amygdala were also evaluated using FSL FIRST masks

(Patenaude et al., 2011). Signal change values were submitted to statistical analyses in SPSS and effects were considered significant at a threshold of $p \leq 0.05$ (two-tailed). We also report results from whole-brain voxelwise analyses at a threshold of $p < 0.005$, cluster minimum = 10 voxels. This threshold was derived from suggested standards for whole-brain comparisons (Lieberman and Cunningham, 2009)

4.2.8 Mediation Analysis

PROCESS software (v.2.11; Hayes, 2013) implemented in SPSS was used to test for the mediating effects of brain function in the association between trauma exposure and symptoms/affective traits. Indirect effects are considered significant when confidence intervals do not overlap zero (Hayes, 2013).

4.3 Results

Trauma and comparison groups were well matched on age, sex, IQ, race, pubertal maturation, annual household income, and movement during the scan (Table 4). One subject was left-handed. Notably, trauma-exposed youth reported lower levels of reward responsiveness (BAS_{rr}), but did not differ on anxiety or depressive symptoms (see Table 4). VAS scores did not differ between groups, $t(28) = 0.314$, $p = 0.76$, suggesting that effects reported are not likely influenced by group differences in state fear/anxiety.

4.3.1 Behavior

Overall task performance: Groups did not differ on overall task accuracy, $t(30) = 1.24$, $p = 0.22$, or RT, $t(22.69) = 1.99$, $p = 0.06$ (Figure 9A), but a trend in group differences was observed such that trauma participants tended to respond faster than the comparison group.

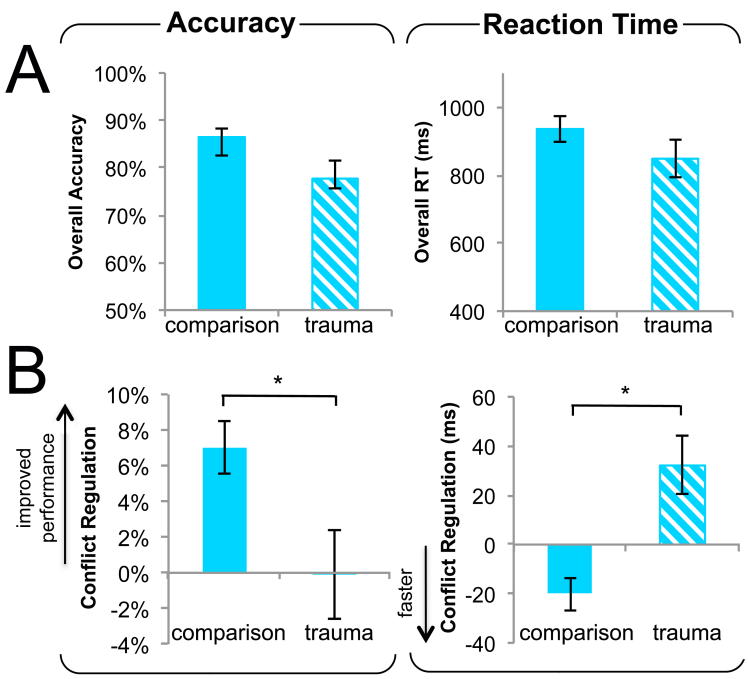

Figure 9. Lower emotion conflict regulatory ability in trauma-exposed youth. (A) No group differences were observed in overall accuracy (left) or reaction time (RT; right). (B) Trauma-exposed youth showed lower ability to regulate emotional conflict. Left: positive values indicate a gain in performance for iI relative to cI trials (iI-cI), Right: negative values indicate faster response for iI relative to cI trials (iI-cI). *$p \leq 0.05$, two-sample *t*-test. Error bars represent standard error of the mean.

Effects of task congruency: As expected, emotional conflict (I-C) induced a slowdown in RT and a reduction in accuracy across the sample, p's < 0.001 (see Table 5). Groups did not differ on conflict interference (I-C; accuracy: $t(30) = 0.9$, $p = 0.37$; RT: $t(30) = 1.77$, $p = 0.086$).

Table 5. Behavioral effects of task congruency

	ACC (%)		RT (ms)	
Trial type	M	SD	M	SD
Congruent (C)	86.91	10.63	859.39	171.18
Incongruent (I)	77.99	15.09	898.19	186.96

Abbreviations: ACC, accuracy; RT, reaction time; mean, M; standard deviation, SD.

Conflict regulation effects: In the comparison group, participants made ~7% fewer errors and improved RT by ~20 ms for repeat conflict trials, a pattern that is consistent with adaptive response to conflict and improved performance. In contrast, trauma participants did not show this

adaptive gain in performance across repeat interference trials. Trauma participants showed no gain in accuracy; in fact they showed a ~35 ms reaction time performance decrement when iI trials were compared to cI trials. Furthermore, adaptation differences between groups were significant for both accuracy, $t(30) = 2.08$, $p = 0.046$, and RT, $t(27.25) = 2.04$, $p = 0.046$ (see Figure 9B). We discovered no evidence indicating that group differences in conflict regulation were moderated by pubertal maturation (accuracy: $F(1, 28) = 0.24$, $p = 0.63$; RT: $F(1,28) = 0.33$, $p = 0.57$).

4.3.2 Aberrant Conflict-Related Amygdala Activity in Trauma-Exposed Youth

Amygdala *a priori* region of interest analysis revealed greater conflict-related activity (I-C) in right, $t(28) = 2.11$, $p = 0.04$, Cohen's $d = 0.79$, and left, $t(28) = 2.23$, $p = 0.03$, Cohen's $d = 0.84$, amygdala in trauma-exposed relative to comparison participants (Figure 10A-C). There were no trauma-by-puberty interactions for left ($F(1, 26) = 0.22$, $p = 0.65$) or right ($F(1, 26) = 1.83$, $p = 0.19$) amygdala responses to conflict. No group differences in amygdala activity were observed during conflict regulation (iI-cI).

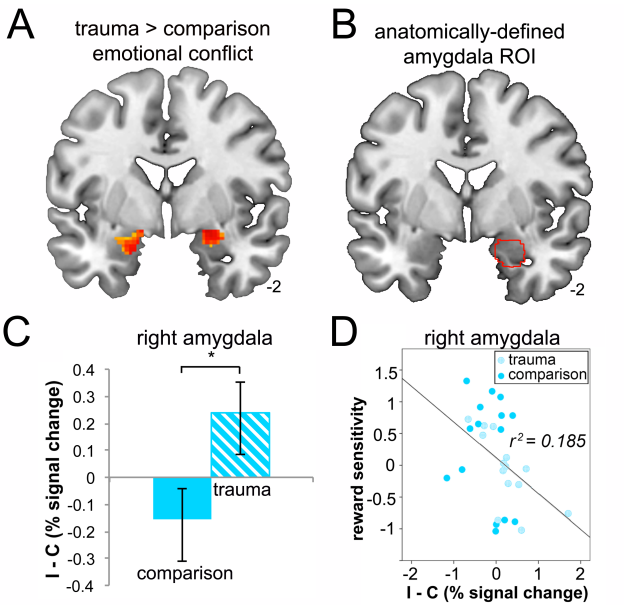

Figure 10. Greater amygdala response to emotional conflict in trauma-exposed youth. Greater conflict-related (I-C) bilateral amygdala activity was observed in trauma relative to comparison participants. Results are displayed for the whole brain at $p < 0.05$ uncorrected (A). The anatomically-defined amygdala region from which average signal was extracted (B) to provide group differences shown in (C). *$p \leq 0.05$ two-sample *t*-test. Higher conflict-related amygdala reactivity across the sample mediated the association between trauma exposure and diminished reward sensitivity (D). Error bars represent standard error of the mean.

4.3.3 Trauma-Exposed Youth Show Abnormal Regulation of the Dorsolateral Prefrontal Cortex

We observed significant small volume-corrected group differences in the left DLPFC during emotional conflict regulation (x = -46, y = 30, z = 38, pFWE = 0.027, Z = 3.10). Relative to comparison youth, trauma participants displayed elevated left DLPFC activity (Figure 11A). Failure of trauma participants to dampen DLPFC activity in iI trials paralleled their inability to improve accuracy and RT during these trials (see Figure 9B,C) and this association was significant: among trauma-exposed youth, higher DLFPC activity during emotional conflict regulation correlated with reduced ability to improve accuracy during iI relative to cI trials, $r(14)$ = -0.585, p = 0.028. Considerate of possible pubertal effects we evaluated the trauma-by-puberty

interaction for left DLPFC response during conflict regulation; the result was non-significant, $F(1, 26) = 1.4$, $p = 0.24$. No differences in pgACC activity were observed between groups.

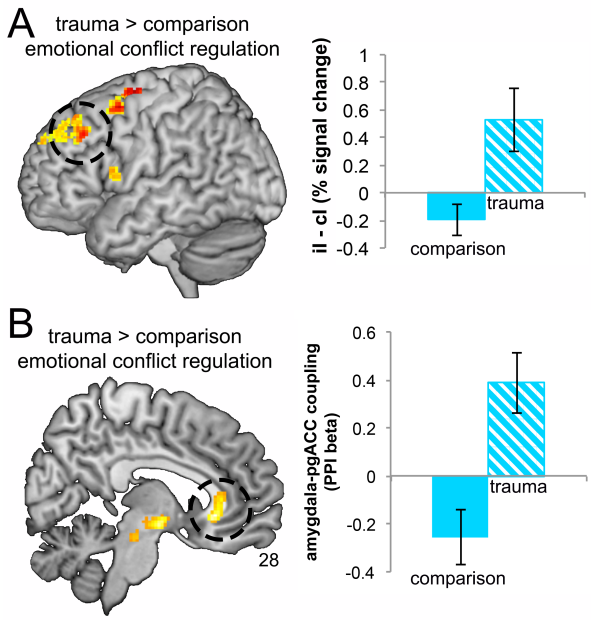

Figure 11. Abnormal regulation of the dorsolateral prefrontal cortex (DLPFC) and disrupted amygdala-pregenual cingulate (pgACC) connectivity in trauma-exposed youth. Increase in DLPFC response is observed in trauma participants (A) and this is accompanied by reduced ability to improve behavioral performance during repeat conflict trials (iI-cI; see Fig.9B). Psychophysiologic interaction functional connectivity analysis showed that only comparison participants showed robust negative connectivity between the amygdala and pgACC during emotional conflict regulation (B). Reduced regulatory connectivity was associated with lower ability of trauma participants to improve reaction time during iI relative to cI trials (iI-cI; see Fig.9B). Whole-brain effects displayed at $p < 0.01$ uncorrected.

4.3.4 Absent Amygdala-Pregenual Cingulate Connectivity in Trauma-Exposed Youth

We observed a significant group difference in amygdala connectivity with the pgACC ($x = 6$, $y = 28$, $z = -4$, pFWE $= 0.016$, $Z = 3.64$) during emotional conflict regulation (iI-cI; see Figure 11B). Extraction of average connectivity strength within this cluster revealed that the group effect resulted from the predicted negative amygdala-pgACC connectivity in comparison participants during conflict regulation, which was absent in trauma-exposed youth. Moreover, less negative amygdala-pgACC FC was associated with reduced ability to improve RT for iI

relative to cI trials, $r(30) = 0.406$, $p = 0.026$. The effect of trauma exposure on amygdala-pgACC connectivity was not moderated by pubertal maturation, $F(1, 26) = 3.58$, $p = 0.07$.

4.3.5 Altered Neural Activity Mediates the Relation Between Trauma Exposure and Deficits in Reward Sensitivity

We evaluated whether neural responsivity and trait RS were correlated within brain regions in which neural function differed between groups (i.e., amygdala, DLPFC, pgACC). We observed significant negative correlations between RS and conflict-related activity in the left, $r(29) = -0.38$, $p = 0.04$ and right amygdala, $r(29) = -0.43$, $p = 0.02$ (Figure 10D). That is, participants with low levels of RS had higher levels of amygdala response to emotional conflict. These effects were driven by variation in the BAS_{rr} subscale (left amygdala: $r(29) = -0.64$, $p < 0.001$; right amygdala: $r(29) = -0.58$, $p < 0.001$), and held when controlling for anxiety and depression symptoms. RS was not correlated with DLPFC activity or amygdala-pgACC connectivity during emotional conflict regulation, p's > 0.1.

We conducted mediation analyses to determine whether conflict-related amygdala reactivity statistically mediated the relationship between trauma-exposure and individual variation in trait RS. A mediation relationship (indirect effects) existed for both the left ($\beta = 0.08$, SE = 0.15, LLCI [lower limit confidence interval] = 0.001, ULCI [upper limit confidence interval] = 0.64) and right amygdala ($\beta = 0.23$, SE = 0.13, LLCI = 0.02, UCLI = 0.56). Direct effects of trauma on RS were not significant, suggesting that amygdala reactivity fully mediated the pathway between trauma exposure and variation in RS. Reversal of this model (trauma exposure→RS→amygdala response) yielded nonsignificant indirect effects (right amygdala: LLCI = -0.03, ULCI = 0.35; left amygdala: LLC: = -0.03, ULCI = 0.28), implying that altered neural response mediates RS but not the reverse.

4.3.6 Exploratory Whole-Brain Results

Results of whole-brain analyses are provided in Table S2 (see the data supplement[3]). Briefly, trauma-exposed showed higher activation to conflict (I-C) than comparison youth in regions of the DLPFC, cerebellum, midbrain, primary and secondary visual areas, cuneus, and precentral gyrus. Trauma-exposed youth also showed higher response during conflict regulation (iI-cI) in primary and secondary visual areas, middle frontal gyrus, precentral gyrus, and preceneus. There were no regions with higher activation to conflict or conflict regulation in comparison youth than in trauma-exposed youth at a $p < 0.005$, 10 voxel minimum threshold.

4.4 Discussion

While research in adults has illuminated lasting neurobiological consequences and deficits in emotion regulation associated with childhood trauma, it is unknown if neural changes are evident in childhood and adolescence, which may serve as a prelude into emotion problems observed in adulthood. The present study tested the hypothesis that childhood trauma exposure alters neural and behavioral systems for detecting and regulating emotional conflict in formative, developmental years. We employed an emotional conflict paradigm in which emotional processing is regulated automatically, and adapted this task for children (see Figure 8). Three main findings emerged. First, compared to their unexposed counterparts, youth with histories of trauma showed greater amygdala reactivity to emotional conflict. Second, trauma-exposed children and adolescents were unable to regulate emotional conflict, indexed by a lack of improvement in accuracy and RT to repeat emotionally incongruent trials. This behavioral deficit was accompanied with a failure to regulate the DLPFC, and an absence of negative regulation-related amygdala-pgACC connectivity in trauma-exposed youth. Third, we found evidence for

[3]http://www.nature.com.proxy.lib.wayne.edu/npp/journal/v40/n5/suppinfo/npp2014311s1.html

trait-brain associations: conflict-related amygdala reactivity was associated with diminished levels of RS.

Engagement of the amygdala is thought to confer preferential processing to emotional stimuli (Vuilleumier et al., 2001) so that potential threats can be rapidly detected and evaluated (LeDoux, 1996). Given this role, heightened amygdala reactivity to emotional conflict observed in trauma-exposed youth may reflect alterations in the neural systems that monitor the environment for biologically salient information. There is substantial evidence that amygdala reactivity is under inhibitory control of medial prefrontal regions (Ochsner and Gross, 2005), particularly pgACC (Maier and di Pellegrino, 2012). Our results indicate that this mechanism is disrupted in children with early life trauma, evidenced by (1) reduced ability to regulate emotional conflict, (2) exaggerated amygdala response to conflict, and (3) absence of effective inhibitory control (i.e., amygdala-pgACC connectivity). While we did not observe group differences in pgACC activation during conflict regulation as previously reported in adults with anxiety disorders (Etkin et al., 2010), we did replicate the finding of lower negative functional connectivity between amygdala and pgACC. It is possible that altered pgACC regional responses observed during adulthood result from presence of psychiatric disorder, or compensation resulting from earlier emerging disruptions in amygdala-pgACC circuitry. We recently reported that resting intrinsic connectivity of the amygdala-pgACC regulatory pathway is altered in youth with histories of trauma (Thomason et al., In Press), suggesting that these deficits are pervasive and are detected even when the individual is not engaging emotion regulatory processes. Similar deficits in top-down control are observed in adults with high levels of trait anxiety (Hare et al., 2008), generalized anxiety disorder (Etkin et al., 2010), and major depressive disorder (Etkin and Schatzberg, 2011). As trauma-exposed youth in the present study were unable to automatically

regulate emotional processing, it is possible that children's early experiences may alter attentional thresholds (indexed by amygdala reactivity) in ways that undermine effective emotion regulation (see review by; Pollak, 2008). Notably, group differences in amygdala response to conflict occurred in the absence of differences in behavioral interference, suggesting that elevated amygdala reactivity represents a latent neural mechanism that enhances the processing of emotional information.

Prior work in healthy adults emphasizes that the DLPFC is online during conflict detection, and is then dampened during resolution (Etkin et al., 2006). DLPFC activity during conflict resolution in trauma-exposed but not comparison youth suggests aberrant engagement of attentional control. Moreover, persistent activation of the DLPFC in trauma participants correlated with observed behavioral deficits, i.e., reduced ability to regulate emotional conflict. It is notable that, similar to trauma-exposed youth in the present study, adults with major depression show increased DLPFC activity during emotional conflict regulation, and an absence of amygdala-pgACC inhibitory control in the same task used here (Etkin and Schatzberg, 2011). However, unlike trauma-exposed youth, depressed adults were not impaired in their ability to behaviorally regulate emotional conflict. In contrast to our findings, DLPFC activity correlated with *better* emotion regulatory ability, suggesting that DLPFC recruitment in depressed adults reflects effortful control in support of emotional conflict regulation (Etkin and Schatzberg, 2011). Altogether, our results suggest that dysfunction of the DLPFC observed in trauma-exposed youth may *interfere* with resolution of emotional conflict, and/or represent inefficient neural resources recruited in an effort to *overcome* this deficit.

A wealth of research shows that early life adversity is one of the strongest predictors of psychopathology (Gilbert et al., 2009). While trauma-exposed youth did not show higher levels

of anxiety or depression, they reported experiencing decreased positive affect in response to rewarding stimuli (i.e., lower levels of BAS_{rr}). This is striking given that diminished RS is emerging as a promising trait marker of disease susceptibility (Bogdan and Pizzagalli, 2006) and severity (Kasch et al., 2002). Deficits in reward processing are particularly relevant for the onset of psychopathology in youth, predicting increases in depressive symptoms (Morgan et al., 2013) and poorer response to evidence-based treatments during adolescence (McMakin et al., 2012). Moreover, changes in reward processing are thought to contribute to key affective and motivational features of anhedonia (Treadway and Zald, 2011).

Our results demonstrate novel associations between early life trauma, deficits in emotion regulation, and variation in RS during formative years. Specifically, trauma in early life predicts reduced RS through altered function of the neural systems that process emotion. This is in line with a growing body of literature linking early life stress to reward processing dysfunction (Bogdan et al., 2013). Lack of positive engagement during childhood and adolescence may contribute to a loss of the normal protection or resilience against traumatic stress. Highlighting the specificity of this effect, no relationships were observed for anxiety or depression, and RS effects held when controlling for these variables.

Study limitations are important to note. First, we tested the effects of trauma in an urban, low-income sample that is predominantly female and African American. One must therefore be cautious when generalizing these effects. Second, sample size was limited, and replication in larger samples is warranted. Third, a high number of participants were excluded due to excess motion and/or low task performance. While this is a situation common in pediatric studies, future studies examining conflict interference in youth should consider shortening the task length, introducing more breaks, or having a researcher remain in the room with the participant to ensure

task compliance and remind them to be still. Fourth, this was a cross-sectional study, and thus it is not possible to determine how variation in neural and behavioral responses during conflict regulation impact long-term outcomes. Longitudinal evaluation will be necessary to determine how specific observations herein relate to vulnerability or resilience to trauma in early life.

In summary, we demonstrate trauma-related perturbations in the neural and behavioral systems that underlie emotional conflict regulation in childhood/adolescence. Trauma-exposed youth showed greater amygdala response to emotional conflict, reduced ability to regulate emotional conflict, failure to engage amygdala-pgACC regulatory circuitry, and ineffective DLPFC engagement. These findings imply a simultaneous heightened sensitivity to conflicting emotional information and a lack of regulatory control over emotion processing in youth who have experienced trauma. This "double hit" is likely to limit the ability of the child to master age-appropriate skills in social and academic domains. We speculate that attentional biases and emotion regulation difficulties may confer elevated risk for psychopathology in youth exposed to trauma. Here, trauma-related changes were detected in systems responsible for the automatic regulation of emotion, in line with emerging evidence that the root of emotion regulatory difficulties in psychopathology might be in more spontaneous forms of emotion regulation (Etkin et al., 2010). Our results show trauma-related changes in neural systems that regulate emotional conflict in youth, offering a potential target for interventions.

CHAPTER 5: CHANGES IN CONNECTOME-LEVEL BRAIN ORGANIZATION AND CONFLICT INTERFERENCE IN YOUTH EXPOSED TO TRAUMA

5.1 Introduction

Information about the wforld is funneled into our brain through sensory organs. Various inputs compete for our attention, and we prioritize and weight these inputs in favor of those most relevant to our goals. The process by which we select amongst competing stimuli is rapid and predominately automatic. Current neurobiological models hold that aberrant filtering, detection, and mapping of salient external stimuli or internal mental events plays a significant role in psychopathology (Menon, 2011). According to these models, increased bottom-up detection of salient events impairs the ability to recruit higher-order brain systems mediating attention and cognitive control. Elevated interference by inappropriately assigned salient information may underlie cognitive dysfunction and emotion regulatory deficits hallmark of several psychiatric disorders.

Childhood trauma exposure is a critical and significant risk factor, associated with ~50% of childhood psychiatric disorders and ~30% of later-onset clinical disorders (Green et al., 2010). A growing body of evidence indicates that disruptions in cognitive processes and their associated neural underpinnings may contribute to the elevated risk in these individuals. These studies show altered perceptual sensitivity and attention control (review by Pollak, 2008), and potentiated neural responses to salient stimuli in individuals who have experienced early adversity or trauma (McCrory et al., 2011, Dannlowski et al., 2012, Herringa et al., 2013b). Hyperactivity of brain regions that detect and enhance biologically-relevant information, such as the amygdala, fronto-insular cortex (comprising the anterior insula and ventrolateral prefrontal cortex), and dorsal anterior cingulate cortex (dACC), are consistently reported findings. Activity in these regions correlates with emotional arousal (Taylor et al., 2003), autonomic activity (Critchley, 2005), and

anticipation of aversive events (Kalisch et al., 2005). Notably, these regions are key nodes of the salience network (SN), an intrinsic connectivity network involved in detecting, integrating, and filtering relevant interoceptive, autonomic, and emotional information (Seeley et al., 2007, Taylor et al., 2009). Prior studies also demonstrate that activity and connectivity within the SN is elevated in adults with major depressive disorder (Hamilton et al., 2012, Manoliu et al., 2013), PTSD (Sripada et al., 2012), and anxiety disorders (Etkin and Wager, 2007). It has been postulated that aberrant function and interaction of the SN may contribute to negative biases in attention and thought inherent in these disorders (Etkin and Wager, 2007, Hamilton et al., 2012). It is possible that functional changes in the SN are a consequence of childhood trauma that also serves to increase psychiatric risk.

Neurobiological investigations have yet to provide clarity regarding functional brain changes that link the experience of childhood trauma with the development of psychopathology. First, the majority of neuroimaging studies are conducted in adults with histories of childhood trauma. Effects observed in adults may reflect secondary compensatory mechanisms rather than those *primarily* associated with trauma. Evaluation of childhood and adolescence, periods proximal to the time of the traumatic experience, may inform understanding of early emergence of neurological traits that are precursors to mental illness. Recent pediatric research has begun to address this gap, revealing heightened sensitivity of SN regions to emotional stimuli in youth who have experienced early adversity or trauma (Maheu et al., 2010, McCrory et al., 2011, Tottenham et al., 2011, White et al., 2012). To our knowledge, SN engagement during interference processing has yet to be examined in a high-risk developmental trauma framework. Second, because most either lack behavioral measures or find no differences in behavior, changes in brain activation may reflect core deficits, compensation, or both. In addition, few

studies link neural changes to psychiatric symptoms, and even fewer examine specific symptom dimensions (e.g., reward sensitivity) which may show greater correspondence with neurobiological variation (Morris and Cuthbert, 2012). Third, we are aware of no pediatric studies that examine the impact of childhood trauma on resting-state functional connectivity of the SN.

Based on prior findings of altered perceptual sensitivity and attention control and heightened neural responses to salient stimuli in adults that experience early trauma, we test the hypothesis that salience processing and functional organization of the SN becomes disrupted in youth - proximal to the traumatic experience. An overactive SN alerting system may contribute to cognitive and socioemotional deficits observed in individuals who have experienced trauma in early life. Tasks that create conflict (i.e., tension between two competing stimuli) provide a reliable behavioral metric of the ability to detect and filter extraneous stimuli, and the degree to which conflicting information interferes with ongoing cognitive processing. Evaluation of neurocognitive function during conflict allows us to measure engagement of SN regions and the relevance of neural changes for behavior. A prior study in women with PTSD (related to interpersonal trauma) found higher responses to interference in the insula, supporting the notion of increased salience detection in the SN in those who have experienced trauma and/or secondary disease symptomology (Bruce et al., 2012). While the amygdala is traditionally thought of in the context of *emotional* conflict, research shows that core SN regions, including the dACC and fronto-insular cortex, track conflict more broadly (Egner et al., 2008, Aupperle et al., 2014).

The fronto-insular cortex (FIC) is considered an integral hub of the brain, regulating information flow across other large-scale brain networks involved in attentional processing and cognitive control (Sridharan et al., 2008). Research shows that the right FIC (rFIC), in particular,

mediates switching between self-directed (e.g., default mode network) and executive control networks (Sridharan et al., 2008), and that this switch has relevance for mental health (Hamilton et al., 2011). The FIC contains a specialized class of neurons with large axons that facilitate rapid relay of control signals to other cortical regions (Cauda et al., 2014), and is thus well suited to initiate control signals. Developmental research shows that the FIC is one of the earliest developing structures in the prenatal period (Afif et al., 2007), and its role as a integral hub of the brain is established within the first years of life (Fransson et al., 2011, Gao et al., 2011). Although the critical role of this structure is evident in early life, there is evidence that large-scale functional brain connectivity continues to undergo significant restructuring throughout childhood (Menon, 2013). The FIC, in particular, shows weak within- and between-network functional connectivity in childhood (Uddin et al., 2011), highlighting its potential source of vulnerability for developmental psychopathology.

The FIC is also an important site of convergence for salient proprioceptive, interoceptive, emotional, cognitive, homeostatic and environmental inputs. Information originating from sensory perceptive regions is received by the amygdala and FIC, extending into mid-posterior insular regions (Cauda et al., 2014). The amygdala and mid-posterior insula are considered part of the extended SN, given evidence from both human neuroimaging (Seeley et al., 2007, Taylor et al., 2009) and primate tract tracing (Mesulam and Mufson, 1982) literatures. Although salience filtering likely occurs at multiple levels of processing, current theory holds that the SN (and the rFIC in particular), triggers a cascade of cognitive control signals, impacting how stimuli are subsequently processed (Menon and Uddin, 2010). This unique role underscores the potential for profound disruptions in cognitive and affective functioning should insular function or connectivity be altered.

Here, we describe research that examined the impact of childhood trauma on connectivity within the SN during rest and neurocognitive function during a face-categorization conflict task. We hypothesized that youth exposed to trauma would show greater behavioral decrements to conflicting stimuli (i.e., higher conflict interference), increased response to conflict in SN brain regions, and increased connectivity within the SN. While we focus on function and connectivity within the SN, we also tested for altered connectivity between the SN and the default mode network (DMN), given the SN's critical role in initiating network switching and prior research showing changes in SN-DMN connectivity in adults with MDD (Manoliu et al., 2013) and PTSD (Sripada et al., 2012). Finally, we evaluated the correspondence between observed variation in the developing connectome and individual variation in positive and negative valence systems. These dispositions are relevant for psychopathology (as outlined by the Research Domain Critiera initiative; Morris and Cuthbert, 2012) and offer insight into the manner of differences observed between groups, aiding interpretation of neural effects as risk or adaptation factors. A self-report measure of trait RS was used to assess the positive valence systems, while validated anxiety and depressive symptom measures were used to assess negative valence systems.

5.2 Methods

5.2.1 Participants

A total of 51 youth, recruited locally through advertisements or child psychiatry clinics (Detroit, Michigan), participated in this fMRI study. Exclusionary criteria included: English as a second language, lower than a 2nd grade reading level, history of brain injury, neurological or movement disorders, or presence of MRI contraindications. Parental informed written consent and child/adolescent assent were obtained prior to participation. The Human Investigation Committee of Wayne State University approved the study protocol.

Both trauma and comparison participants were recruited based on high sociodemographic risk. Prior research shows that trauma frequency is extreme among African Americans living in impoverished areas (nearly 90%; Gillespie et al., 2009). Moreover, minority, urban residents are nearly two times more likely to develop emotional psychopathology following trauma exposure (Kessler et al., 1995, Gillespie et al., 2009, Goldmann et al., 2011). Despite this population's apparent increased susceptibility to mental illness following trauma, little research has examined trauma and its neural correlates in high-risk, urban residents.

IQ was evaluated using the Kaufman Brief Intelligence Test (KBIT v.2; Kaufman and Kaufman, 2004a). Pubertal maturation was assessed using Tanner staging (Marshall and Tanner, 1968). Following prior work (Forbes et al., 2009), participants were categorized as pre/early (Tanner stages 1-2) or mid/late pubertal (stages 3-5). Resting-state data from 22 participants have been reported previously (Thomason et al., 2013). Face categorization conflict task data presented here have never previously been reported, however data from an analogous emotion-categorization stroop task in 29 participants included here have previously been described (Marusak et al., 2014a; tasks counterbalanced for order of presentation). Although we did not conduct diagnostic testing or exclude individuals with attention-deficit/hyperactivity disorder (ADHD), data from initial study screening noted 3 trauma and 1 comparison participant for potential ADHD-like behavior. Two participants were on psychotropic medications: one trauma participant was taking atomoxetine and sertraline, and one comparison participant was taking trazodone, guanfacine, methylphenidate, sertraline, and clonidine. Follow-up analyses excluding the two participants on medications yielded no changes to observed effects.

5.2.2 Trauma and Clinical Measures

Utilizing parent and youth reports, youth participants who experienced at least one trauma indicated on the Children's Trauma Assessment Center Screen Checklist (source: Michigan Trauma Assessment Center) were categorized as 'trauma'. Number and type of endorsed traumas are provided in Table 6. Participants with movement exceeding 4 mm or 3 rotational degrees (n = 12; 3 trauma, 9 comparison), conflict task accuracy <50% (n = 4; 1 trauma, 3 comparison), or errors in behavioral data collection (n = 2; 1 trauma, 1 comparison) were excluded from analyses. Therefore, all data are reported for 14 trauma-exposed and 19 age-, sex-, and IQ-matched comparison youth.

Table 6. Participant demographics by group

	Trauma (n = 14)	Comparison (n = 19)
Age, m (SD)	12.61 (2.11)	12.06 (2.66)
Sex (female), n (%)	10 (71.4)	15 (78.9)
Pubertal development, n (%)		
Pre/early pubertal (Tanner stages 1-2)	5 (35.71)	8 (42.1)
Mid/late pubertal (Tanner stages 3-5)	9 (64.29)	11 (57.9)
IQ, m (SD)	100.14 (13.17)	104.29 (14.34)
Race/Ethnicity, n (%)		
African American	5 (35.71)	9 (47.37)
Caucasian	3 (21.43)	8 (42.11)
Hispanic	2 (14.29)	0
Biracial	1 (7.14)	1 (5.26)
Not reported	3 (21.43)	1 (5.26)
Household Annual Income, n (%)		
Less than $40,000	10 (71.43)	9 (47.37)
$40-60,000	2 (14.29)	4 (21.05)
$60-80,000	1 (7.14)	3 (15.79)
Over $80,000	0	3 (15.79)
Not reported	1 (7.14)	0
Type of Trauma Endorsed, n (%)		
Physical abuse	2 (14)*	0*
Neglectful home environment	2 (14)*	0*
Exposure to domestic violence	7 (50)*	0*
Exposure to any other violence not already	7 (50)*	0*

identified		
Multiple separations from parent or caregiver	2 (14)*	0*
Sexual abuse or exposure	3 (21)*	0*
Anxiety Symptomology (SCR), m (SD)	19.38 (13.97)	13.38 (8.94)
Depressive Symptomology (CDI), m (SD)	2 (2.56)	2.26 (3.01)
Reward Sensitivity (BAS, z-scores), m (SD)	-0.2 (0.59)	0.07 (0.94)
Reward responsivitiy	17.21 (1.58)	18 (2.09)
Fun seeking	12.07 (1.9)	12.44 (2.67)
Drive	10.43 (2.47)	11 (2.56)
Motion During Cognitive Conflict Task[a], m (SD)		
Translational mean movement	0.05 (0.03)	0.05(0.03)
Rotational mean movement	3.44 (4.01)	4.01 (5.16)
Translational RMS	0.05 (0.03)	0.05 (0.03)
Rotational RMS	0.06 (0.03)	0.06 (0.04)
Translational max excursion	0.47 (0.5)	0.44 (0.26)
Rotational max excursion	0.45 (0.36)	0.46 (0.29)
Motion During Resting-State Scan[a], m (SD)		
Translational mean movement	0.15 (0.12)	0.18 (0.13)
Rotational mean movement	0.12 (0.12)	0.14 (0.1)
Translational RMS	0.1 (0.06)	0.11 (0.05)
Rotational RMS	0.001 (0.001)	0.001 (0.001)
Translational max excursion	0.66 (0.43)	0.85 (0.39)
Rotational max excursion	0.47 (0.31)	0.57 (0.28)

**Indicates group comparison is significant at $p \leq .05$. Chi-square tests were used for sex, race/ethnicity, income, and trauma-type comparisons; two-sample t-tests for age, psychopathology, and motion comparisons; Mann-Whitney U for income. [a]Translational (x, y, z) movement is reported in mm; rotational, in degrees. Parenthetical values given by totals or means represent percentages and SDs, respectively. Abbreviations: standard deviation, SD; mean, m; Intelligence Quotient, IQ; Screen for Child Anxiety Related Emotional Disorders, SCR; Children's Depression Inventory, CDI; Behavioral Activation Subscale of the BIS/BAS, BAS; root-mean-square (head position change), RMS.*

Individual variation in positive and negative valence systems was assessed, as outlined by the Research Domain Criteria initiative (Morris and Cuthbert, 2012). Variation in negative valence systems were measured using two validated self-report measures of anxiety and depressive symptoms: the 41-item Screen for Child Anxiety-Related Emotional Disorders (SCR; Birmaher et al., 1997) and the 10-item Children's Depression Inventory (CDI; Saylor et al., 1984). The 20-item Behavioral Inhibition and Activation Scales (BIS/BAS; Carver and White,

1994) was used to measure variation in positive valence systems. Trait RS was conceptualized as the Behavioral Activation (BAS) component of the BIS/BAS, following prior work (Garner et al., 2012, Marusak et al., 2014a). Scores for each of the three BAS subscales (representing different aspects of reward function: reward responsiveness, fun seeking, drive) were converted to z-scores and averaged to form RS, an overall index of reward function. RS data were not available for one comparison participant. A visual analog scale (VAS) was used to obtain an average rating of fear/anxiety during the MRI visit (repeat measures at 30-minute intervals) as previously described (Thomason et al., 2013).

5.2.3 Imaging Data Acquisition

BOLD fMRI data were obtained using a 3 Tesla Siemens Verio scanner equipped with a 12-channel head coil (MRI Research Center, Wayne State University). Conflict task and resting-state scans utilized the same acquisition parameters. Both used T2*-weighted echo-planar imaging with TR: 2000 ms, TE: 25 ms, matrix: 220 × 220, flip angle: 90°, voxel size: 3.44 × 3.44 × 4 mm, and acquired twenty-nine axial slices. High-resolution anatomical images were acquired using a T1-weighted 3D MP-RAGE sequence with TR: 1680 ms, TE: 3.51 ms, orientation: axial, matrix: 384 × 384, 176 slices, flip angle: 90°, voxel size: 0.7 × 0.7 × 1.3 mm.

5.2.4 Participant Head Motion

Motion poses a significant challenge for fMRI research (Power et al., 2012). The following three steps were taken to address motion-related artifacts. (1) High-movement resting-state frames (exceeding 3.3 mm or 2.2 rotational degrees) were removed using ArtRepair software (http://cibsr.stanford.edu/tools/human-brain-project/artrepair-software.html). This threshold resulted in the removal of no more than 10% of frames (i.e., <18 out of 180) per participant. (2) Average movement across the scan, root-mean-square (rms) head position

change, and maximum excursion (frame-to-frame displacement) were calculated and averaged for translational (*x, y, z*) and rotational (*roll, pitch, yaw*) movement directions. Participants with excess movement were excluded from the study sample.

For remaining participants, motion was well within accepted standards (< 1.5 mm rms; Fair et al., 2012). Motion parameters were compared between groups using 2-tailed independent samples *t*-tests for both fMRI experiments. (3) Motion-related signal (6 realignment parameters with another 6 parameters representing their first order temporal derivatives) was removed with covariate regression analysis before computing SN and DMN connectivity. Signals from white matter and cerebral spinal fluid were also regressed out using anatomical component correction (aCompCor; Behzadi et al., 2007, Chai et al., 2012).

5.2.4 Experimental Paradigms and Procedures

5.2.4.1 Conflict task: During fMRI, participants underwent a face categorization conflict task adapted from Egner et al. (2008).The task consisted of 163 presentations of happy or fearful facial expression photographs, overlaid with the words "FEMALE" or "MALE" to create categorically congruent and incongruent stimuli (see Figure 12A). Participants were instructed to identify the gender of the face stimuli with a button press response, while trying to ignore the task-irrelevant gender word stimuli. Stimuli were presented for 1,000 ms, with a varying interstimulus interval of 2,000-4,000 ms (mean = 3,000 ms), in a pseudorandom order, counterbalanced across trial types for expression, word, response button, and gender. The original task (Egner et al., 2008) utilized adult face stimuli. Here, we adapted the task for children by utilizing an established set of child and adolescent face stimuli (Egger et al., 2011), minimizing the complex relations inherent in adult face stimuli (Marusak et al., 2013). Stimuli were presented with EPrime software v.2.0 (Psychology Software Tools, Inc., Pittsburgh, PA)

during fMRI scanning and displayed on a back-projection screen that was viewed by the participants via a mirror attached to the head coil. Task duration was 12:46. Participants with poor task performance (<50% accuracy) or errors in behavioral data collection were not included in the study sample. For remaining participants, task accuracy was fair (mean = 86.4%, SD = 9.92%). Reaction time (RT) was unavailable for one participant due to errors in data collection, and this participant was therefore not included in RT analyses but was retained in all other analyses for completeness.

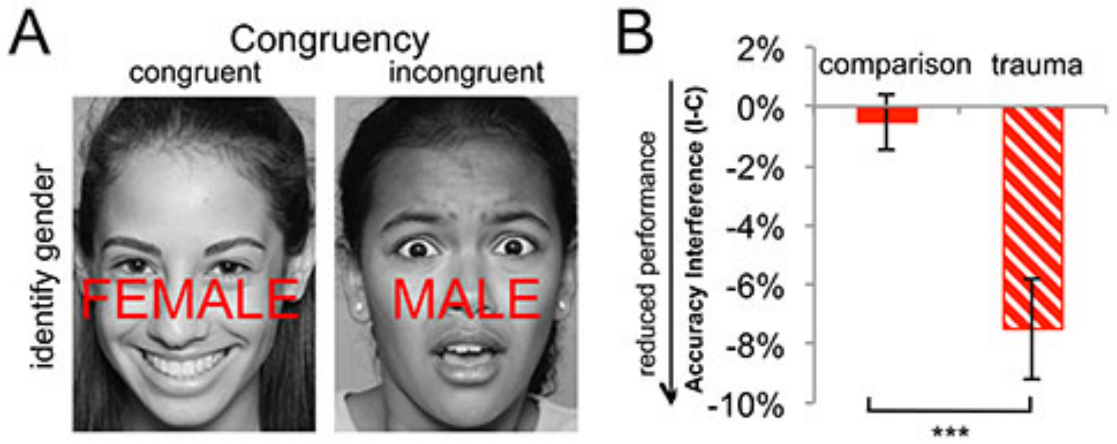

Figure 12. (A) Face categorization conflict task and (B) group differences in conflict interference. Participants were instructed to identify the underlying face gender (male or female) while ignoring an overlying gender word ('MALE' or 'FEMALE'). Trials varied such that distracter words either matched ("congruent") or conflicted ("incongruent") with the underlying face. Trauma-exposed youth show a greater loss of accuracy for incongruent relative to congruent trials (I-C). Negative values indicate a loss in performance. ***$p < 0.001$, two-sample *t*-test. Error bars represent standard error.

5.2.4.2 Resting-state paradigm: Following the conflict task, participants underwent a 6-minute resting-state paradigm. Participants were asked to lie quietly in the scanner with their eyes closed for the duration of the scan.

5.2.5 Imaging Data Analysis

5.2.5.1 Preprocessing: BOLD fMRI data were processed using SPM8 software (Statistical Parametric Mapping; http://www.fil.ion.ucl.ac.uk/spm/) implemented in MATLAB (MathWorks, Inc., Natick, MA). The first four image volumes were excluded to allow for signal equilibration effects. Preprocessing steps included: (i) slice-time correction, (ii) image realignment, (iii) spatial transformation to the Montreal Neurological Institute (MNI) template using the participant-specific transformation parameters created by fitting mean functional images to the single reference EPI standard template (in SPM). Data were not resampled during normalization, thus retained the native resolution (3.44 x 3.44 x 4 mm) for subsequent analysis. (iv) Images were then spatially smoothed with a Gaussian kernel (6 mm full width at half maximum [FWHM] for task data; 8 mm FWHM for resting-state data).

5.2.5.2 Conflict task: A 128-second temporal high-pass filter was applied to the data, and temporal autocorrelation was estimated using a first-order autoregressive model. Two independent participant-level models were created in the context of a general linear model to examine effects of (1) conflict and (2) conflict regulation. In the first model, separate regressors for the stimulus events (convolved with a canonical hemodynamic response function) were created for incongruent (I) and congruent (C) trials. For the second model, trial types were broken down based on the preceding trial type: regressors were created for postcongruent incongruent trials (cI), postincongruent incongruent trials (iI), postcongruent congruent trials (cC), and postincongruent congruent trials (iC). All participant-level models included regressors of no interest corresponding to the six motion parameters, and modeled error and posterror trials separately. Participant-level contrasts isolated (1) conflict-related neural activity by subtracting congruent from incongruent trials (I-C), and (2) conflict regulation by subtracting post-congruent incongruent trials from post-incongruent incongruent trials (iI-cI). The contrast iI-cI isolates

activity during conflict trials for which behavior differs by virtue only of priming induced by conflict demands of the previous trial type (i.e., previous trial is either congruent or incongruent). Engagement of the conflict system on the preceding trial should ready the system for reengagement. Group-level random-effects two-sample *t*-tests were used to test for group differences in neural activity during (1) cognitive conflict (I-C), and (2) conflict regulation (iI-cI).

5.2.5.3 Resting-state: Connectivity analyses were performed using the CONN fMRI functional connectivity toolbox (version 12.1; www.nitrc.org/projects/conn). Resting-state fMRI volumes were submitted to seed-based connectivity analyses to assess connectivity within the SN, and between the DMN and SN. First, seed time series data were extracted from SN and DMN masks comprising 6 mm radii spheres centered at Montreal Neurological Institute (MNI) coordinates of peak-valued loci as determined by group averaged independent components analysis of intrinsic functional networks identified in an independent N = 65 pediatric sample (Thomason et al., 2011): (41, 21, -5), (-2, 23, 33), and (-46, 15, -5) for the SN, and (5, -53, 13), (-2, 57, -18), and (52, -63, 26) for the DMN, see Figure S2 in the online version of this paper[4]. Correlation estimates controlled for estimated translational and rotational motion as well as a white matter and cerebral spinal fluid nuisance time course. A band-pass filter was applied to investigate low-frequency correlations (between 0.01 and 0.1 Hz; Van Dijk et al., 2010). Pearson bivariate correlation coefficients were calculated between average time courses in the SN seed region mask and all other voxels of the brain. Group-level random-effects two-sample *t*-tests were used to test for group differences in intrinsic connectivity of the SN, and connectivity between the DMN and key SN regions.

[4]http://www.sciencedirect.com.proxy.lib.wayne.edu/science/article/pii/S2213158215000741

5.2.5.4 Regions of interest: To examine the relevance of intrinsic SN connectivity for neural engagement during a cognitive task, all between-group effects were considered within SN regions known to be recruited in the conflict task: (i) bilateral amygdala (left, x = -30, y = -6, z = -14; right; x = 32, y = 0, z = -12), (ii) dACC (x = 2, y = 32, z =31) and (iii) rFIC (x = 40, y = 30, z = -7) using coordinates derived from our prior work (Etkin et al., 2006, Egner et al., 2008), and (iv) a mid-posterior insula region (x = -38, y = -13, z = -8) that showed higher responses in adults with PTSD during a similar task (Bruce et al., 2012). Regional masks (10 mm radii spheres) were created around each peak and then intersected with a gray matter mask. Group differences were examined within each region separately using a threshold of $p < 0.05$, small-volume family-wise error (FWE) corrected.

5.2.5.5 Exploratory whole-brain results: Between-group whole-brain effects of I-C are also reported at a threshold of $p < 0.005$, cluster minimum= 10 voxels. This threshold was derived from suggested standards for whole-brain analyses (Lieberman and Cunningham, 2009).

5.2.6 Relations Among Measures

5.2.6.1 Correlations among measures: Average signal change/connectivity strength was extracted from peaks of group difference (4 mm radii spheres) and plotted for visualization, and/or submitted to 4 planned Pearson correlation analyses in IBM SPSS v.22 to evaluate correspondence amongst brain activation, connectivity, task performance, and symptom severity. Specifically, we tested for relations between insula connectivity and (i) insula reactivity to conflict, (ii) behavioral response to conflict, and (iii) RS, and (iv) between insular and behavioral responses to conflict. All coordinates provided in this report are given in MNI convention.

5.2.6.2 Mediation analysis: PROCESS software v.2.11 (Hayes, 2013) implemented in SPSS was used to test for the mediating effects of insula-SN connectivity in the association

between trauma exposure and RS. We focused on RS due to prior research documenting relations between neural effects related to trauma exposure and this symptom dimension (Bogdan et al., 2013, Marusak et al., 2014a). Relations between neural connectivity and symptoms related to negative valence (anxiety, depression) were also examined. The mediation model assumes significant relations between trauma exposure and connectivity, and between connectivity and RS. Connectivity values were extracted from the insula region showing increased SN connectivity in trauma-exposed youth. We then evaluated the correspondence between connectivity strength and RS. This approach uses bootstrapping, and indirect effects are considered significant when confidence intervals do not overlap zero (Hayes, 2013).

5.3 Results

Trauma and comparison groups were matched on age, sex, pubertal maturation, IQ, race, annual household income, and movement during both fMRI experiments (Table 6). Two trauma participants were left-handed. Groups did not differ on RS, anxiety, or depressive symptoms (Table 6). Participants also did not differ on ratings of state anxiety obtained via visual analog scale (VAS), $t(31) = 0.08$, $p = 0.94$. Lack of differences between groups in state and trait mood symptoms suggests that observed effects are not influenced by group differences in fear and anxiety.

5.3.1 Greater Behavioral Decrements to Conflict in Youth Exposed to Trauma

Consistent with the conflict effect, incongruent trials (relative to congruent trials) caused significant accuracy decreases and slowing in reaction time (RT) across the sample, accuracy: $t(32) = 3.26$, $p = 0.003$; RT: $t(31) = 3.8$, $p = 0.001$. Relative to comparison youth, trauma participants showed a greater impairment in accuracy for incongruent vs. congruent trials (I-C), $t(31) = 3.8$, $p < 0.001$ (Figure 12B). Specifically, conflict trials caused ~8% loss in accuracy in

trauma-exposed youth, while performance was relatively consistent across incongruent and congruent trials in comparison youth. This is in line with the notion that trauma-exposed youth show greater behavioral interference by task-irrelevant distracters. Breakdown by trial type (I,C; see Figure S2 in online data supplement) showed that this group difference was driven by lower accuracy in trauma participants during incongruent rather than congruent trials (group x trial-type interaction, $F(1,31) = 14.42$, $p = 0.001$). No group differences in RT interference or conflict regulation were observed, p's > 0.17 (Table 7). Groups did not differ on overall task accuracy or RT (p's > 0.29; Table 7).

Table 7. Behavioral performance on the conflict task by group

	Trauma (n = 14)	Comparison (n = 19)	*p*-value
Overall ACC (%), m (SD)	84.3% (8.87%)	87.96% (10.58%)	0.3
Overall RT (ms), m (SD)	802.24 (200.31)	879.54 (216.55)	0.31
Conflict Regulation ACC (%)[a], m (SD)	-1.85% (9.08%)	1.3% (6.8%)	0.18
Conflict Regulation RT (ms)[a], m (SD)	27.37 (61.33)	-2.56 (61.63)	0.26

[a]Conflict regulation is defined by postincongruent incongruent trials minus postincongruent congruent trials (iI-cI). For ACC, lower numbers indicate greater performance decrements for incongruent relative to congruent trials. For RT, higher numbers indicate greater slowdowns for incongruent relative to congruent trials. P-values derived from two-sample t-tests. Abbreviations: mean, m; standard deviation, SD; reaction time, RT; accuracy, ACC; milliseconds, ms.

5.3.2 Trauma-Exposed Youth Show Elevated Insula Reactivity to Conflict

We observed greater rFIC response to conflict (I-C) in trauma-exposed relative to comparison youth, $x = 32$, $y = 34$, $z = -10$, $Z = 2.98$, $pFWE = 0.031$, Figure 13A. Across the sample, higher rFIC reactivity was associated with greater performance decrements to conflict (I-C), $r(33) = -0.375$, $p = 0.032$. A similar pattern was observed in left mid-posterior insula; trauma-exposed youth showed higher response to conflict ($x = -34$, $y = -10$, $z = 10$, $Z = 3.89$, $pFWE = 0.002$) which was associated with greater performance decrements, $r(33) = -0.433$, $p =$

0.012, Figure 13B. Amygdala and dACC SN regions did not show group differences in responses to conflict, and neural activity did not differ between groups during conflict regulation (iI-cI). Exploratory whole-brain effects of I-C are provide in Table S2 (see online data supplement). Briefly, trauma-exposed youth showed higher response to conflict (I-C) in the FIC, putamen, inferior parietal lobe, and sensorimotor areas; comparison youth showed higher response to conflict in the inferior parietal lobe.

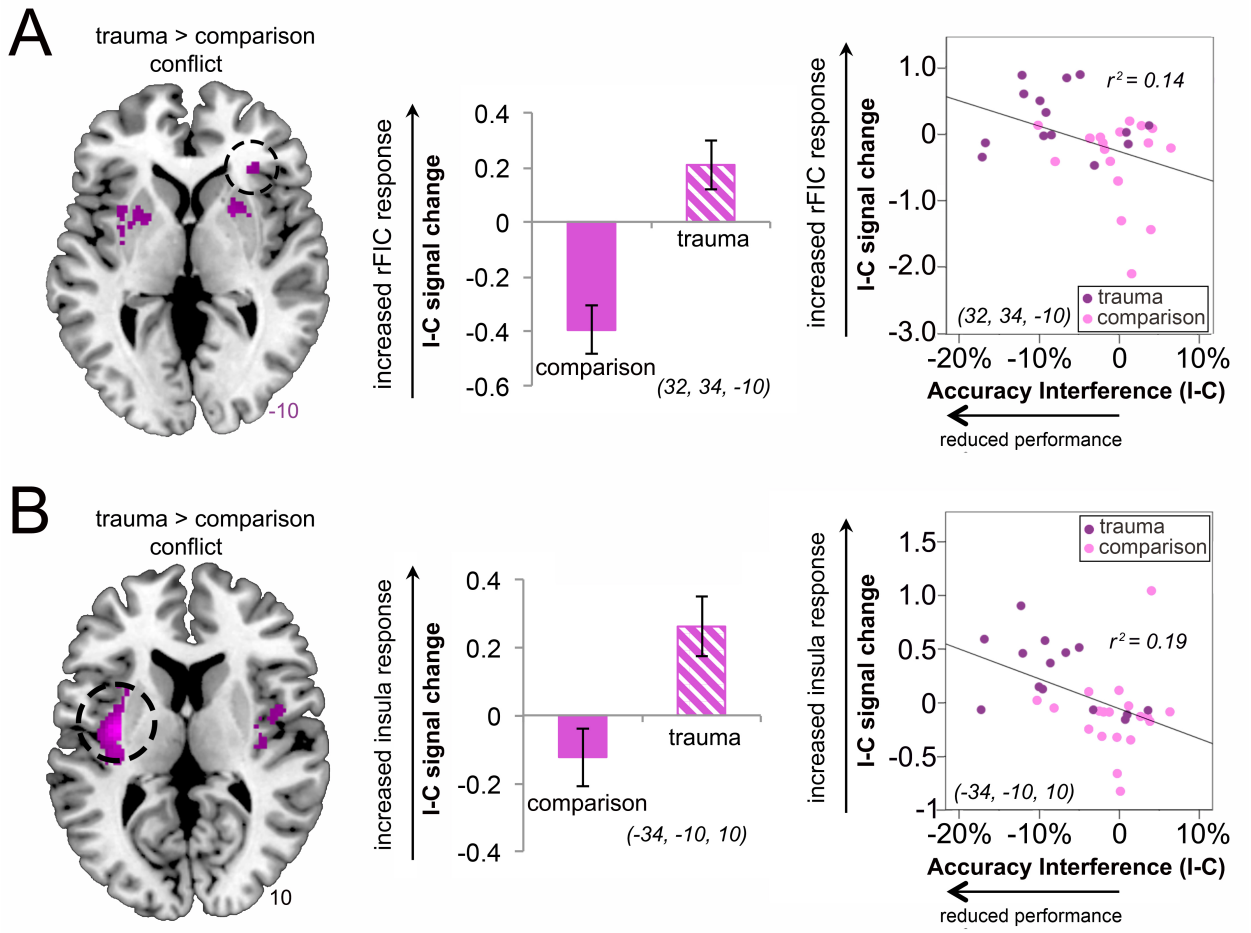

Figure 13. Trauma-exposed youth show greater (A) right fronto-insular cortex (rFIC) and (B) left mid-posterior insula response to conflict, that correlates with greater performance decrements. Fronto-insular response to conflict is exaggerated in trauma-exposed youth, especially those that demonstrate large interference values (incongruent-congruent trials [I-C]). Clusters are significant at *pFWE* < 0.04, small-volume corrected. X, Y, Z coordinates are given in MNI convention. Error bars represent standard error.

\5.3.3 Aberrant Salience Network (SN) Connectivity in Trauma-Exposed Youth

Relative to comparison participants, trauma-exposed youth demonstrated increased SN connectivity within the left amygdala (x = -28, y = -6, z = -18, Z = 3.13, $pFWE$ = 0.046; Figure 14A) and left middle insula (x = -28, y = 8, z = -16, Z = 3.98, $pFWE$ = 0.011; Figure 14C), and reduced SN connectivity in the right dACC (x = 4, y = 36, z = 34, Z = 3.14, $pFWE$ = 0.044; Figure 14B). Increased SN connectivity in the left middle insula was associated with higher rFIC response to conflict (I-C; Figure 14D), $r(33)$ = 0.516, p = 0.002, and greater performance decrements to incongruent trials, although the latter effect was a non-significant trend, $r(33)$ = -0.335, p = 0.057. SN connectivity across the sample is presented in Figure S3 (see online data supplement).

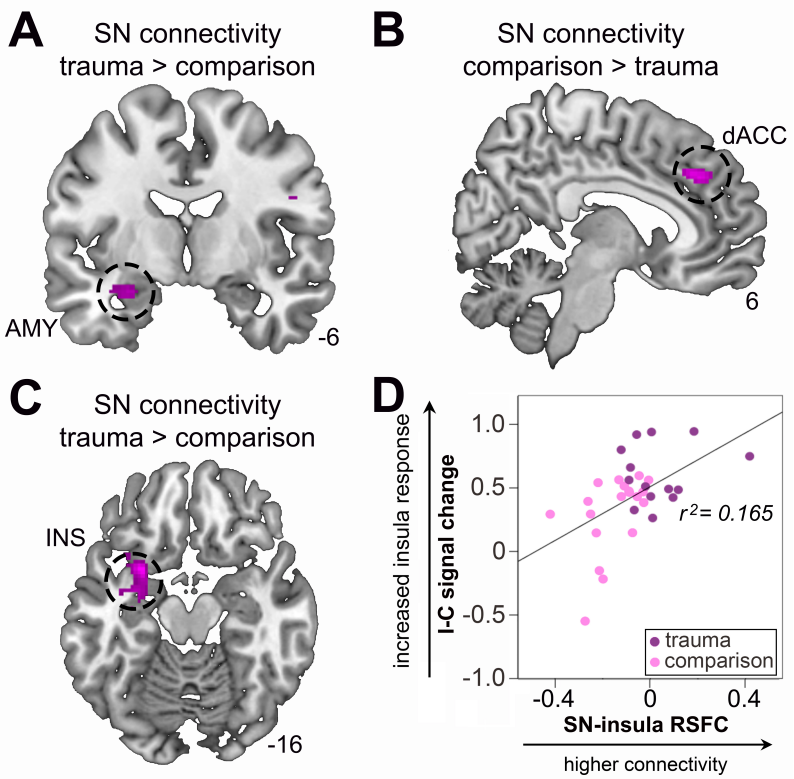

Figure 14. Group differences in salience network (SN) connectivity. Trauma-exposed youth show increased amygdala (AMY), A, decreased dorsal anterior cingulate cortex (dACC), B, and increased middle insula (INS), C, to SN signal covariance. Increased SN connectivity within the left insula (x = -28, y = 8, z = -16) was associated with greater right fronto-insular (x = 32, y = 34,

z = -10) response to conflict, D. Results are significant at $pFWE < 0.05$, small-volume corrected. X, Y, Z coordinates are given in MNI convention. Error bars represent standard error.

5.3.4 Altered Connectivity within the Salience Network (SN) is Associated with Variation in Trait Reward Sensitivity (RS)

Driven by prior work showing relationships between depressive symptoms and altered SN connectivity in the insula (Manoliu et al., 2013), and by our recent work showing associations between trait RS and altered function of emotional conflict neural systems in trauma-exposed youth (Marusak et al., 2014a), we tested associations between SN-insula connectivity and RS. Strength of SN connectivity was extracted from the peak of the insula region that showed higher connectivity in trauma-exposed youth. We then tested for associations between connectivity in this region and RS across the sample. We observed that higher SN to left insula connectivity was associated with diminished RS, $r(32) = -0.373$, $p = 0.036$. Mediation analyses showed that the association between trauma exposure and RS was mediated by SN-insula connectivity (β = -0.4, standard error (SE) = 0.21, lower limit confidence interval = -0.89, upper limit confidence interval = -0.007; see Figure 15). Neither anxiety nor depressive symptoms were related to SN connectivity in the insula (p's > 0.27).

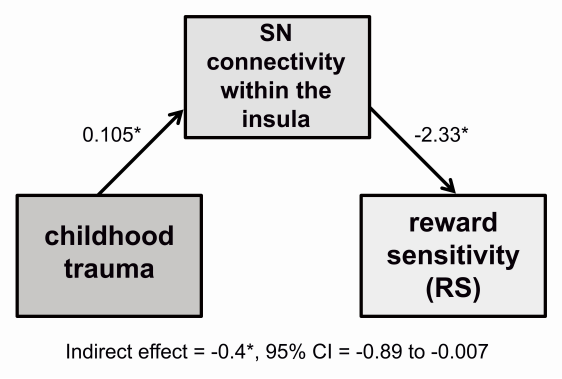

Figure 15. Salience network (SN) connectivity within the insula mediates the relationship between trauma exposure and reward sensitivity (RS). Unstandardized regression

coefficients and bias-corrected 95% confidence interval (CI) for the indirect effect from a bootstrap-mediation analysis. Specifically, trauma exposure led to diminished RS through increased SN connectivity within the insula. *$p < 0.05$.

5.3.5 Aberrant Connectivity between the Salience Network (SN) and Default Mode Network (DMN) in Trauma-Exposed Youth

Next, we evaluated connectivity between the SN and the DMN. As shown in Figure 16, trauma-exposed youth showed reduced DMN to SN connectivity, particularly in the dACC (x = 2, y = 26, z = 40, Z = 4.05, $pFWE$ = 0.011). DMN connectivity across the sample is presented in Figure S3 (see online data supplement).

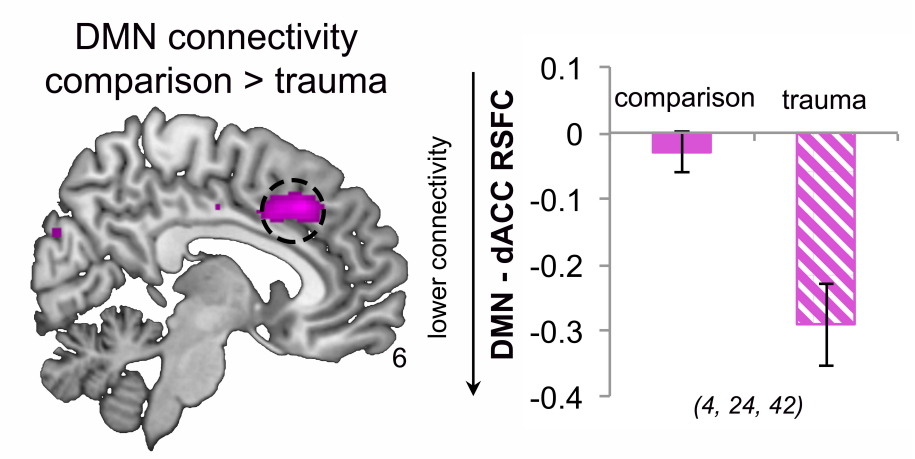

Figure 16. Altered connectivity between the salience network (SN) and the default mode network (DMN) in trauma-exposed youth. Trauma-exposed youth show lower DMN connectivity with the right dorsal anterior cingulate cortex (dACC), a key SN node. Resting-state functional connectivity is depicted as Fisher-transformed r values. Results are significant at $pFWE$ = 0.011, small-volume corrected. Coordinates are given in MNI convention. Error bars represent SEM.

5.4 Discussion

Contemporary neurobiological models suggest that inappropriate assignment of saliency to external stimuli or internal mental events leads to aberrant interactions within and between

large-scale neurocognitive networks, and plays a significant role in several psychiatric disorders (Menon, 2011). Recent research in adults supports this conceptualization (e.g., Sripada et al., 2012, Manoliu et al., 2013). However, it is unknown if changes are evident in the brain *prior* to the emergence of clinically significant symptoms; these may underlie vulnerability. The present study is the first to link early life trauma exposure – a major predisposing factor for the development of psychopathology – to dysfunctional architecture of large-scale neurocognitive networks in a sample of high-risk urban youth. We demonstrate increased SN connectivity within the insula in trauma-exposed youth that has cognitive repercussions: increased SN connectivity corresponds with suboptimal brain and behavioral responses during a conflict task. Further, we demonstrate that altered SN connectivity is associated with individual variation in positive valence systems. That is, higher SN connectivity within the insula was associated with lower RS. These results suggest that enhanced salience detection, diminished sensitivity to reward, and connectome-level brain changes may contribute to later cognitive and affective deficits observed in individuals who have experienced trauma. A schematic representation of the overarching framework is provided in Figure 17.

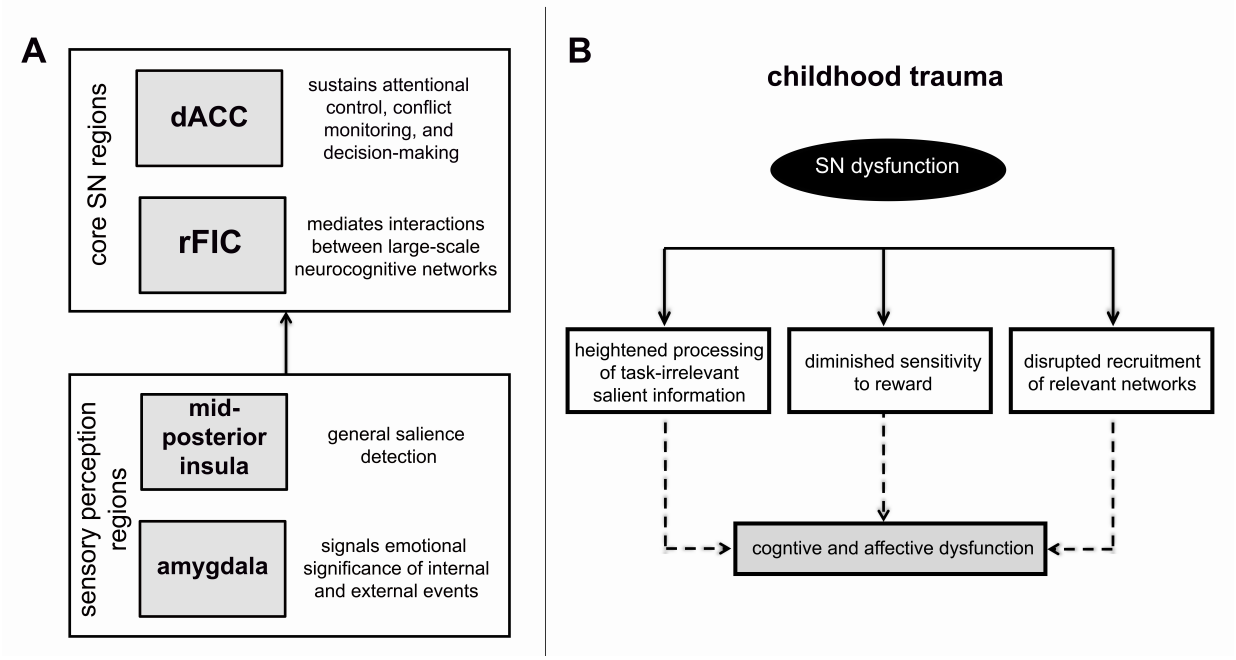

Figure 17. Schematic representation of (A) salience network (SN) organization and (B) model of SN dysfunction in youth exposed to trauma. (A) The normal function of the SN is to detect relevant internal and external cues among myriad inputs. Ventral sensory pathways, including the amygdala and mid-posterior insular regions, feed forward stimulus information to core SN regions (i.e., right fronto-insular cortex (rFIC) and dorsal anterior cingulate cortex (dACC)). Engaged rFIC may influence dominance in other neurocognitive systems that are relevant for goal-directed cognitive processes (e.g., conflict regulation). This is consistent with suggestions that the FIC is an integrative hub that filters how sensory inputs are further processed (Menon and Uddin, 2010). (B) The present results demonstrate that childhood trauma, a major predisposing factor for psychopathology, is associated with altered function and connectivity of the SN. Fitting with conceptual models of posttraumatic stress disorder (Patel et al., 2012) and major depressive disorder (Hamilton et al., 2012), changes within the SN may lead to a cascade of events that increase risk for cognitive and affective dysfunction hallmark of these disorders. Our results suggest that altered salience processing may underlie the link between early life trauma and development of psychopathology. Specifically, we observed greater performance decrements to conflict trials, which corresponded with increased fronto-insular responses in trauma-exposed youth. We also observed altered connectivity within the SN, and between the SN and the default mode network (DMN). Network-level disruptions may therefore underlie the observed sub-optimal brain and behavioral responses during the conflict task in trauma-exposed youth. Finally, strength of SN connectivity within the insula was associated with reduced reward sensitivity, an affective trait emerging as an important risk/resilience factor in the aftermath of early life trauma exposure.

Trauma-exposed youth show elevated rFIC and left mid-posterior insula response to conflict relative to comparison youth. The latter is striking given that a similar left mid-posterior

insula region (x = -37, y = -21, z = 12) was shown to be hyperreactive in women with PTSD during an emotional interference task analogous to the paradigm employed in the current study (Bruce et al., 2012). In that study, participants were instructed to indicate whether two nonemotional house stimuli in the horizontal axis were the same or different, while ignoring distracting emotion-face pairs in the vertical axis. Posterior insular regions are thought to interact with anterior fronto-insular regions to modulate autonomic reactivity to salient stimuli. Once a salient stimulus is detected, the rFIC initiates attentional control signals and facilitates network switching. In the current study, higher reactivity of both rFIC and mid-posterior insula to conflict was associated with greater performance decrements. This suggests that increased salience detection in fronto-insular regions may interfere with the ability to recruit higher-order cognitive systems that are necessary for task execution, or cause inappropriate engagement of higher-order systems to task-irrelevant, conflicting stimuli.

We observed greater contributions of the insula and amygdala to the SN in trauma-exposed youth. This finding is consistent with prior research in adults with PTSD (Sripada et al., 2012). Our results thus extend earlier observations and suggest that strengthening of connectivity within a network that detects salient external and internal events begins proximally to the traumatic experience, during youth. Moreover, we observed that patterns detected at rest corresponded with those observed during a neurocognitive task. Specifically, SN connectivity was higher in the insula, which was more responsive to conflict in trauma-exposed youth. These converging results support the notion that elevated SN activity is a pervasive phenomenon that may affect these individuals across a variety of contexts. These data are also fitting with prior behavioral reports of sustained attention to extraneous stimuli in youth who have endured early life trauma (Pechtel and Pizzagalli, 2011). Inappropriate assignment of salience to mundane

events could interfere with ongoing cognitive or affective processes by biasing attentional resources. For instance, elevated salience detection could diminish a child's ability to focus on the task at hand, or to regulate emotional responses.

Trauma and comparison groups did not differ on demographic factors measured, allowing us to compare the effects of trauma exposure in groups that were similar in sociodemographic risk. Given also that the trauma group did not present with marked clinical symptomology relative to the comparison group, observed neural changes may represent either risk or adaptation to adverse early environments. Our results support the former. Specifically, we observed that youth showing the most aberrant pattern of insula-SN covariance (e.g., elevated connectivity) showed diminished levels of RS. RS is an affective trait that may contribute to the emergence of stress-related psychopathology during adolescence (Bogdan et al., 2013) and as such, dysfunctional SN connectivity within the insula may represent a neural substrate of increased risk. Longitudinal follow-up planned in this sample may allow for further evaluation of this potential mechanism.

Our analyses indicate that SN connectivity is lower in the dACC in trauma-exposed youth. This is in contrast to the observed strengthening of SN connectivity in the amygdala and middle insula. This result suggests reduced reliance on a core SN region (i.e., dACC), with a concomitant increase in regions thought to carry salient interoceptive, homeostatic and emotional information to core SN nodes (i.e., amygdala, middle insula). In line with a recent conceptual framework (Menon, 2011), we demonstrate a link between aberrant connectivity of the rFIC within the SN and aberrant connectivity between the SN and DMN, a network involved in self-referential processing (Buckner et al., 2008). Prior research shows that altered connectivity of large-scale neural networks has relevance for mental health. For instance, depressed adults show

altered connectivity within the SN's rFIC and strength of connectivity relates to symptom severity (Manoliu et al., 2013). The network-level brain changes observed in the present study therefore hold implications for psychiatric risk. Altered network connectivity may also provide an avenue for intervention. For example, a recent study showed that transcranial magnetic stimulation (TMS) was capable of attenuating aberrant network connectivity in depressed individuals by modulating interactions between networks (Liston et al., 2014), an encouraging result.

Study limitations warrant mention. First, sample size was limited, and replication in larger samples is warranted to improve generalizability of the observed effects. Thus, results are presented as preliminary, but highlight important neural and behavioral differences in trauma-exposed youth in an understudied population of urban-dwelling, minority youth with a high stress burden. Next, we lack information about the onset (age) and duration of trauma experienced by these study participants. While retrospective analyses show that trauma onset and type relate to distinct emotional outcomes (English et al., 2005), prior studies also document nonspecific effects of these variables on outcomes (Arata et al., 2007, Collishaw et al., 2007) and some suggest that disentangling unique effects may result in overly narrow interpretations (Green et al., 2010). Future studies should also consider obtaining trauma documentation beyond self- and parent-report measures (e.g. positive forensic investigation by Child Protective Services). Next, it is unclear why groups did not differ in levels of internalizing symptomology (anxiety, depression, RS). This may be because both groups were drawn from the same sociodemographic risk community, and did not differ on IQ, race, or income. Comparing groups with similar backgrounds provides a unique opportunity to isolate effects of trauma in the context of high-risk youth. However, future research might consider comparing trauma groups

with and without diagnoses or presence of elevated symptom levels. Additionally, since this research is cross-sectional, we cannot examine which, if any, of these participants will go on to develop stress-related clinical disorders. While this work provides the first characterization of SN integrity and conflict systems in trauma-exposed youth at high risk for clinical disorders, future longitudinal work is needed to evaluate disease trajectories. Finally, while we observed a significant relationship between trauma and SN function, other factors are likely to influence and be influenced by SN function, such as age and gender. Effects of additional factors and their potential interactions with trauma is important for future work that may seek to develop personalized interventions to promote optimal outcomes in children who have experienced trauma.

5.4.1. Conclusions

The present results demonstrate a compelling pattern of SN dysfunction, particularly in the insula, in youth exposed to trauma. We show that trauma-exposed youth are more susceptible to interference during conflict and this correlates with higher fronto-insular responses to conflict, and increased tethering of the insula to the SN. We therefore provide evidence of trauma-related changes across multiple domains of neural function, and show that the observed effects have relevance for behavior. Our data also suggest a direct link between connectome-level brain organization and specific symptom dimensions associated with psychiatric risk. In particular, increased insula engagement in the SN was associated with diminished RS, an affective trait that is emerging as a critical risk/resilience factor in the aftermath of stress. Overall, these results support the notion that childhood trauma exposure is associated with disrupted saliency processing at the level of large-scale neural networks. Our findings are preliminary, but may aid the formulation of hypotheses about neural processing differences that result from significant

traumatic life events. Further research will be needed to advance discovery in this area to development of behavioral interventions, a much-needed direction for follow-up work. For instance, mindfulness, cognitive training, and neural stimulation have potential to quell overactive SN nodes and alter the functional connectome (Lutz et al., 2013, Liston et al., 2014).

CHAPTER 6: CONCLUSION

6.1 Summary of findings

A series of studies is outlined in this dissertation that reveal neural impacts of childhood trauma in children and adolescents (youth). Relative to matched comparison youth, trauma-exposed youth show altered neural response and FC in core emotion processing brain regions, e.g., amygdala, pgACC/sgACC, insula. Similar FC alterations are detected at-rest, when youth are not actively engaging in emotion processing (see Chapters 3 and 5). The major findings of these studies are summarized and their potential implications are further discussed, below.

Given the central role of the amygdala in emotion processing, we began with a study evaluating structure and function of the amygdala (Chapter 2). We found that the amygdala was more responsive to salient social cues in youth carrying a particular variant of the oxytocin receptor gene (A allele), and that this effect was amplified in youth with higher levels of early stress exposure. Expanding on these findings, we found that exposure to childhood trauma was associated with altered resting-state FC of the amygdala with sgACC, a critical emotion regulation pathway (Chapter 3). Trauma-exposed youth also showed heightened amygdala response to emotional conflict, and lower amygdala-pgACC FC during emotion regulation (Chapter 4). In Chapter 5, we expanded our view to test how trauma impacts functional neural responses, as well as FC within and between relevant networks. We found heightened neural response to conflict in the insula, a key salience network region, that corresponded with altered FC of the salience network.

As discussed in Chapter 1, emotion systems are critical for lifelong health and wellbeing. Disruptions in these systems are hallmark of several common debilitating psychiatric disorders, including anxiety, depression, and PTSD. Here, we discovered neurobehavioral alterations in

youth who have experienced trauma, one of the most potent risk factors for psychopathology, during a time that marks the beginning of a high-risk period for onset of psychiatric disorders (late childhood/early adolescence; Angold et al., 1998). Thus, observed neurobehavioral changes may serve as potential biomarkers of risk for later psychopathology.

Importantly, we find that neurological changes associated with trauma exposure were also associated with specific behavioral and symptom dimensions. In particular, trauma-exposed youth were less able to regulate emotional conflict (Chapter 4) and were more susceptible to distracting emotional cues (Chapters 2 and 5). Further, we found that neurobehavioral alterations were associated with RS rather than NA. In particular, youth with higher neurobehavioral impairment reported lower levels of RS, a finding that is further considered, below. Overall, these findings support the clinical and behavioral relevance of observed neurological changes.

Research into the neurobehavioral consequences of trauma in youth is motivated by the strong and pervasive link between childhood trauma and psychopathology (Kessler et al., 2010). Empirically based interventions that are capable of diminishing risk and/or circumventing development of psychopathology are badly needed for the 35 million US children affected (Child and Adolescent Health Management Initiative; 2012). We believe that the research described in this dissertation is an important first step towards this goal, and outline future directions and potential avenues for intervention, below.

6.2 Future research directions

One important future direction will be to identify which neural changes precede onset of disorder and which are associated with resiliency. Here, we attempted to address this by linking observed neurobehavioral changes to symptom levels, and found that youth with the most aberrant neurobehavioral patterns reported lower RS. However, longitudinal research is needed

to address whether these changes predict onset of disorder. Longitudinal research is also needed to identify potential mediators and buffers of risk, e.g., genetic variants, expressed emotion in the family, and neighborhood context. Longitudinal research may also provide important clues about the role of puberty in the pathophysiology of trauma-related psychopathology. This may provide greater mechanistic insight into the increased prevalence of emotional disorders in females relative to males that emerges after the onset of puberty (Angold and Costello, 2006).

Another layer of complexity that will be better addressed by longitudinal research is trajectories of brain maturation. Structural neuroimaging studies have found that trajectories of neurobiological change may be more predictive of outcomes than a single measurement at one time point (Shaw et al., 2006). Within-individual trajectories, only accessible in longitudinal studies (Kraemer et al., 2000), should therefore allow for better characterization of developmental deviations related to trauma exposure. Trajectory analyses can reveal shifts in the rate, timing (i.e., ages) or shape (e.g., inverted U, quadratic) of a developmental trajectory, and the impact of different characteristics of the trauma (e.g., age of onset, chronicity). Longitudinal research will provide a more complete understanding of how genetic and environmental factors influence brain, behavior, and affective experience in youth.

Future studies with larger samples will be able to more fully address the interaction between genes and childhood trauma (GxE). In Chapter 2, we found that a common polymorphism in the oxytocin receptor was associated with altered structure and function of the amygdala in youth. We also found evidence for a GxE effect, such that the combination of higher levels of environmental adversity and the *OXTR* A allele was associated with the greatest neurobehavioral dysfunction. These findings are consistent with a stress diathesis GxE model, which holds that the environment is necessary to unmask an underlying genetic susceptibility

(Goforth et al., 2011). However, stress diathesis is not the only GxE model, and some of our prior findings are more in line with a bioecological GxE model. In contrast to stress diathesis, the bioecological model predicts greater heritability in the presence of *advantageous* proximal environments (Bronfenbrenner and Ceci, 1994). For example, we previously found that gray matter volume of hippocampus and sgACC varied with BDNF genotype only in youth without histories of trauma exposure. No effect of BDNF on corticolimbic volume was evident in trauma-exposed youth (Marusak et al., 2015b), which may reflect the restriction of genetic disparity by a suboptimal environment. Taken together, it is apparent that the pattern of GxE may differ based on the brain area or gene/molecular system of interest (e.g., oxytocin vs. BDNF). Multiple GxE models should be considered in the study of environmental adversity.

6.3 Reward sensitivity as a potential marker of stress susceptibility

We found that neurobehavioral alterations associated with trauma exposure were exaggerated in youth with low levels of RS. There is a rich theoretical background purporting that low RS is a vulnerability factor for the development of affective disorders (see Bogdan et al., 2013). Research shows that reward processing deficits are particularly relevant for the onset of psychopathology during adolescence, predicting increases in depressive symptoms (Morgan et al., 2013), greater illness severity and suicidality (Gabbay et al., 2015), and poorer response to evidence-based treatments (McMakin et al., 2012). Moreover, changes in reward processing are thought to contribute to key affective and motivational features of anhedonia, a cardinal symptom of depression (Treadway and Zald, 2011). Our findings suggest that stress-induced changes in RS could be a potential marker of susceptibility to affective disorders.

An emerging body of research supports the notion that reduced RS is a promising mechanism underlying the link between stress exposure and emotional psychopathology

(Bogdan and Pizzagalli, 2006). In experimental animals, chronic stress can diminish reward-seeking behavior and weaken preferences for sucrose solutions (Pryce et al., 2004, Willner, 2005). Research suggests that these effects are mediated by stress-induced changes in the HPA axis. In humans, variation in HPA axis activity and response has been linked to functional and structural differences in striatal and other corticolimbic regions that are involved in reward processing (see Bogdan et al., 2013). Consistent with this, stress-related HPA axis activation has been shown to directly alter function of dopamine (Piazza et al., 1996, Pascucci et al., 2007), a key neuromodulator in the reward system.

At first glance, it seems counterintuitive that trauma-related changes in prototypical fear and anxiety regions (e.g., amygdala, insula) are associated with self-reported RS rather than NA – which is more typically associated with feelings of fear and anxiety. As we saw in Chapter 1, the amygdala and insula are key nodes of the salience network, involved in biasing both positive and negative emotional processing. The amygdala and insula have direct projections to the ventral striatum, which are thought to modulate learning and stimulus-response valuation of potential rewards (see review by Haber and Knutson, 2010). Over time, heightened sensitivity to emotionally laden materials could result in diminished engagement of approach- and reward-related circuitry in trauma-exposed youth. In addition, repeated stress diminishes dopamine signaling within the striatum (Gambarana et al., 1999) and amygdala (Inglis and Moghaddam, 1999), which could also suppress reward behavior. A recent study in mice demonstrated that repeated stress in childhood led to increased risk for anxiety- and depression-related behaviors in adulthood, and this effect was mediated by changes in dopamine D3 receptor expression and altered function in the amygdala and striatum (Seo and Kuzhikandathil, 2015).

The link between childhood trauma and RS is further supported by recent neuroimaging research. Individuals exposed to childhood trauma report elevated symptoms of anhedonia and show diminished reward-related responses in the basal ganglia. These effects are present in adolescence and even decades later, into adulthood (Dillon et al., 2009, Hanson et al., 2015). Intact hedonic capacity and robust neural reward-related circuitry, in contrast, are thought to be protective against depressive symptoms in the aftermath of stress (Nikolova et al., 2012). Emerging data suggest that link between childhood trauma and mood disorders may be unique to traumatic stress experienced before ~age 10 (e.g., Hanson et al., 2015). Consistent with this, a recent study using data from a nationally representative US survey found that trauma during childhood, but not during infancy or adolescence, conferred significant risk for depression in adolescent girls (Marshall, 2016). Altogether, these findings suggest that reward systems might be particularly susceptible to traumatic stress experienced during childhood. It has yet to be determined if these changes are related to, or result from, heightened saliency processing.

To be a marker of stress-related vulnerability, neurological changes associated with low RS should predict subsequent increases in symptomology. To test this, we brought back a subset of youth (N = 21) from the research described in this dissertation. Controlling for baseline levels, low RS predicted higher internalizing, affective and PTSD problems two years later, r's > 0.5, p's < 0.02, as reported by the caregiver (Child Behavior Checklist, Parent Report; CBCL-PR; Achenbach, 1991). Additionally, we found that neural variation associated with low RS – namely, increased amygdala (Chapter 4) and insula response to conflict (Chapter 5), and increased insula-salience network FC (Chapter 5) - continued to predict low RS two years later, r's < -0.6, p's < 0.05. These findings lend preliminary support to the notion that low RS and associated neurological variation are potential markers of risk during the transition into

adolescence. This is especially important given that initial episodes of depression are likely to occur during this developmental transition (Lewinsohn et al., 1994, Kessler et al., 2001), conferring greater risk for depression in adulthood (Lewinsohn et al., 1999). Moreover, low RS during the transition into adolescence predicts later depressive disorder (Pine et al., 1999, Joiner et al., 2002, Forbes and Dahl, 2010).

6.4 Potential implications for intervention

Identifying cross-cutting dimensions that transect an array of psychopathologies, such as RS, could improve detection of at-risk youth and also aid the design of targeted interventions in early life, prior to disease onset. If these findings are replicated in separate samples and in a longitudinal framework, low RS would be an important marker of risk and potential treatment target. This is significant because current empirically supported treatments (e.g., cognitive behavioral therapy) do not adequately resolve RS-related deficits among youth with emotional disorders (Kennard et al., 2006). Thus, interventions that enhance engagement in pleasant activities (e.g., behavioral activation; Jacobson et al., 2001) or teach regulatory strategies for sustaining positive affect (e.g., Positive Affect Stimulation and Sustainment; McMakin et al., 2011) may be beneficial for youth with low RS.

Identified neurobehavioral changes associated with trauma exposure could provide additional avenues for intervention. In particular, we found heightened response in amygdala and fronto-insular cortex to distracting emotional information in trauma-exposed youth. Increased neural response was associated with lower behavioral performance, suggesting that heightened neural processing of emotional materials may be detrimental to cognitive functioning. Thus, strengthening top-down neural pathways involved in attentional orienting and regulation of emotional processing may be a useful remediation strategy for trauma-exposed youth. One such

strategy is attention bias modification treatment, which involves repeated training of attention allocation away from negative stimuli and toward neutral stimuli. This strategy is proven effective in reducing symptoms of anxiety (meta-analysis by Hakamata et al., 2010). There may be other ways to intervene early in the emotion detection process, at the level of salience network processing. For instance, explicit learning of an emotion regulation-related skill (e.g., suppression, cognitive reappraisal) might render that skill more automatic after sufficient practice and mastery (Gyurak et al., 2011). At least some of these interventional strategies might be implemented via hand-held or online training platforms that are age-appropriate, engaging, and easily accessible (e.g., Hardy et al., 2015).

A complementary intervention approach would be to reduce bottom-up signaling in the salience network, a network that monitors internal and external stimuli for emotional salience (Taylor et al., 2009). This might be achieved through mindfulness based interventions (e.g., meditation; Marchand, 2014) or real-time fMRI neurofeedback training (Paret et al., 2014), which can dampen activity in amygdala and insula. Pharmacotherapies can also have inhibitory effects on amygdala and insula activity, including the widely used selective serotonin reuptake inhibitors (Simmons et al., 2009, Godlewska et al., 2012). Given the link between individual variation in oxytocin neurobiology and amygdala responsivity observed in Chapter 2, pharmacotherapies that target the oxytocin system may be fruitful. In fact, intranasal administration of oxytocin is actively being investigated in adults for its utility in preventing the development of stress-related psychopathology and augmenting treatment response. Initial findings in this area in recently traumatized adults are variable, however, suggesting that greater understanding of doses and of interacting neuromodulatory systems (e.g., serotonin, dopamine) is needed (Taylor et al., 2014, Olff et al., 2015).

Importantly, not all youth with histories of trauma reported low RS. This raises the subject of personalized interventions. Greater attention to dimensional symptom measures (e.g., RS, NA) can aid with the identification of at-risk youth, and suggest the most appropriate interventions. A one-size-fits all approach to intervention is not likely optimal, given that not all trauma-exposed youth will develop an emotional disorder, and some will develop other forms of psychopathology, e.g., substance abuse, ADHD. In this research, we focus on emotion processing systems for their central role in core components of emotional disorders. Evaluation of attentional and cognitive control pathways may unearth markers of externalizing or other types of psychopathology that are associated with childhood trauma.

Since neurobehavioral changes were evident in the trauma group regardless of RS levels, aberrant salience network functioning may represent a more broadband effect of vulnerability that is present even in those who do not yet report affective changes (e.g., low RS). Epidemiological studies show that neurobehavioral changes typically associated with NA in childhood (e.g., hypervigiliance, attentional deficits) often precede onset of RS-related symptomology in adolescence (Silk et al., 2012). Thus, interventions that target salience network systems may be useful for preventing subsequent development of low RS in at-risk youth.

6.5 Different dimensions of childhood trauma

In the research described in this dissertation, childhood trauma was broadly defined to include different types of exposures, i.e., abuse, violence, and neglect. There is reason to not be too narrow, as different exposures seem to carry similar risk (Green et al., 2010). In addition, childhood adversities often co-occur and most individuals exposed to childhood trauma have experienced multiple types of trauma (Kessler et al., 2010, McLaughlin et al., 2012). Thus, constraining trauma types to narrow categories may result in findings that are not generalizable.

However, emerging research suggests the presence of two unique dimensions of childhood adversity that likely have different effects on neural development (McLaughlin et al., 2014). These are threat/victimization and deprivation/neglect. These represent abnormal environmental input (e.g., violence and abuse) and a lack of typical species-expected input (e.g., caregiver absence, neglect), respectively.

In our urban Detroit sample, the most common forms of trauma reported were domestic and community violence. This pattern is consistent with prior research in urban populations (Gillespie et al., 2009, Crusto et al., 2010), and suggests that observed neurobehavioral alterations may be more closely associated with early threat exposure rather than deprivation/neglect. Indeed, neglect and caregiver separation were relatively rare in this sample, endorsed less frequently than both physical and sexual abuse. Neurobehavioral alterations related to early deprivation may be better addressed by an ongoing and complementary line of research in orphanage-reared youth, as described in the Introduction (see also Tottenham, 2012). Such research benefits from the fact that the neglect/deprivation ends when stable families adopt the institutionalized youth, thus creating a sample of at-risk youth with variable lengths of exposure. We believe that there is great value in understanding the neurobehavioral correlates of trauma in urban youth, who represent a different population with different types of adversities (violence, abuse) and sociodemographic factors. According to a national survey, violence and abuse are also exceptionally pervasive, affecting more than 50% of US youth (Finkelhor et al., 2013). Urban youth are disproportionately burdened by victimization and associated negative outcomes (Gillespie et al., 2009, Lowe et al., 2014), providing an enriched sample at high risk.

In recognition that deprivation and threat likely have unique impacts on neural development (McLaughlin et al., 2014), we will narrow our focus to threat/victimization

(violence and abuse exposure) in future research. To enhance the generalizability of findings, we will not exclude youth who have experienced other dimensions of trauma. Rather, we will simultaneously measure and control for neurobehavioral effects related to neglect/deprivation.

In addition to type of trauma assessed, it is important to consider the selection of the matched comparison group. A comparison group drawn from different sociodemographic backgrounds makes it difficult to disentangle effects of trauma exposure from chronic environmental stress (e.g., neighborhood quality, socioeconomic status). Here, we chose to match comparison and trauma-exposed groups on sociodemographic variables that are known to influence neurobehavioral function (e.g., age, sex, race, income), or control for them in analyses. This approach attempts to equate environmental stressors between groups to better isolate effects related to trauma exposure. For further discussion of partcipant selection strategies, see review by (Thomason and Marusak, 2016).

The threat/victimization vs. deprivation/neglect framework put forth by McLaughlin and colleagues suggests that early threat exposure leads to increased amygdala sensitivity to salient emotional stimuli. Over time, elevated corticolimbic sensitivity will result in stronger representations of conditioned fear, weakening input of ventromedial prefrontal cortex (vmPFC; encompassing pgACC/sgACC) – a structure critical for emotion regulation. Reduced vmPFC recruitment over time may lead to accelerated pruning, resulting in reduced vmPFC thickness and/or lower vmPFC-amygdala FC observed here. This framework is consistent with our findings, but further imply that lower ability to modulate learned fear is a potential mechanism linking childhood trauma to emotional psychopathology. This neurobehavioral mechanism has not yet been tested in the context of childhood trauma.

6.6 Cognitive flexibility

Results of this dissertation imply a simultaneous heightened sensitivity to salient emotional information and a lower ability to flexibly regulate emotion processing in trauma-exposed youth. This pattern might reflect an adaptation that derives from an adverse rearing environment, allowing for rapid detection of associations between environmental cues and prediction of imminent threat. Adaptive functioning requires, however, that when environmental cues no longer predict adverse outcomes, it is essential to quickly update this learning and suppress fear responding. Cognitive inflexibility and abnormal persistence of learned fear may play a role in increasing risk for psychopathology. This is consistent with behavioral observations of trauma-exposed youth, who demonstrate rigid, inappropriate expression of learned fear (De Bellis and Zisk, 2014). Reduced ability to flexibly adjust learned fear, at the level of brain and/or behavior, may be a core marker of risk for pathology.

One way to model fear learning in the laboratory is with Pavlovian fear conditioning, which involves the pairing of a previously innocuous cue (conditioned stimulus; CS+) with an aversive outcome (unconditioned stimulus, US). After repeated pairings with the US, presentation of the CS alone begins to elicit a conditioned fear response. Research in both humans and rodents show that the amygdala is critical for the acquisition and expression of conditioned fear (LeDoux, 2000).

Learned fear is not immutable. Ways to flexibly adjust learned fear involve learning new associations, but in different ways. One is extinction, which involves the omission of an aversive outcome. Extinction learning occurs after repeated presentations of the CS in the absence of the US, forming a new memory that competes with the fear memory trace (Quirk and Mueller, 2008). The reduction of fear following extinction learning depends on vmPFC suppression of

amygdala output. Importantly, extinction learning and its recall are context dependent, requiring contextual modulation via the hippocampus to trigger vmPFC-amygdala extinction circuitry (Quirk and Mueller, 2008). This is in contrast to fear learning, which is context independent. As such, stress-related abnormalities within the extinction circuitry that results in lower contextual modulation (via hippocampus), hyper-response to threat cues (via amygdala), or deficient fear modulation (via vmPFC) could lead to a behavioral phenotype of persistent overexpression of fear. These components can be teased apart in a fear- and extinction-learning paradigm.

Another way to modify learned fear is with reversal learning, which involves the pairing of a cue or context with the *opposite* outcome. For example, a cue previously associated with a negative outcome is now associated with a positive outcome. Though fear reversal is less understood than fear extinction, there is evidence to suggest that vmPFC is also involved in signaling 'safety' or positive value for stimuli previously associated with an aversive US. The amygdala and striatum, in contrast, track the stimuli that predict the US (see Schiller and Delgado, 2010). Reversal of a context, but not a cue, appears to depend on the hippocampus (Levy-Gigi et al., 2011). An advantage of a reversal-learning paradigm is that it can test whether trauma-related changes are specific to the modulation of negative stimuli, or generalize to both positive and negative. Similarly, deficits in reversal of contexts vs. cues should provide insight into whether hippocampal-dependent contextual processing is affected.

Research in adults with trauma exposure and stress-related psychopathology (e.g., PTSD, anxiety, depression) points to deficits in both extinction recall (typically tested 24 hr following extinction learning; Milad et al., 2009) and reversal learning (Levy-Gigi and Richter-Levin, 2014). Behavioral deficits are accompanied by changes in the brain, particularly altered hippocampal-vmPFC FC, a pathway critical for the contextual modulation of learned fear. There

is also evidence for amygdala hyper-reactivity (Etkin and Wager, 2007), suggesting that amplified fear response may be a downstream consequence of impaired fear modulation. While we found similar functional neural changes here in trauma-exposed youth, the ability to modulate learned fear has yet to be tested as a core mechanism altered by early trauma in youth.

We plan to expand on these findings in future work to test the flexible modulation of learned fear in trauma-exposed youth. Knowledge of the underlying neurobehavioral mechanisms altered by early trauma should provide critical insight into the link between early trauma and later psychopathology. Findings in this area have potential to inform the development of early interventions. For instance, poor contextual recall of extinction could motivate the development of remediation strategies that strengthen hippocampal-dependent learning processes. A deficit in extinction but not in reversal learning could imply that interventions that involve learning salient, new associations (e.g., a reward) to reduce fear might be superior to those utilizing an omission strategy (e.g., exposure-based therapy).

REFERENCES

(2012) 2011/12 National Survey of Children's Health. Child and Adolescent Health Measurement Initiative

Achenbach TM (1991) Manual for the Child Behavior Checklist/4-18 and 1991 Profile: Department of Psychiatry, University of Vermont.

Adolphs R (2001) The neurobiology of social cognition. Current opinion in neurobiology 11:231-239.

Adolphs R (2010) What does the amygdala contribute to social cognition? Annals of the New York Academy of Sciences 1191:42-61.

Afif A, Bouvier R, Buenerd A, Trouillas J, Mertens P (2007) Development of the human fetal insular cortex: study of the gyration from 13 to 28 gestational weeks. Brain structure & function 212:335-346.

Alim TN, Charney DS, Mellman TA (2006) An overview of posttraumatic stress disorder in African Americans. Journal of clinical psychology 62:801-813.

Amat JA, Bansal R, Whiteman R, Haggerty R, Royal J, Peterson BS (2008) Correlates of intellectual ability with morphology of the hippocampus and amygdala in healthy adults. Brain and cognition 66:105-114.

American Psychiatric Association (2000) Diagnostic and statistical manual of mental disorders: Fourth edition, text revision - DSM-IV-TR: American Psychiatric Association.

American Psychiatric Association (2013) Diagnostic and statistical manual of mental disorders: Fifth edition, DSM-5.

Amunts K, Kedo O, Kindler M, Pieperhoff P, Mohlberg H, Shah NJ, Habel U, Schneider F, Zilles K (2005) Cytoarchitectonic mapping of the human amygdala, hippocampal region

and entorhinal cortex: intersubject variability and probability maps. Anatomy and embryology 210:343-352.

Andreasen NC, Flaum M, Swayze V, 2nd, O'Leary DS, Alliger R, Cohen G, Ehrhardt J, Yuh WT (1993) Intelligence and brain structure in normal individuals. The American journal of psychiatry 150:130-134.

Angold A, Costello EJ (2006) Puberty and depression. Child and adolescent psychiatric clinics of North America 15:919-937, ix.

Angold A, Costello EJ, Worthman CM (1998) Puberty and depression: the roles of age, pubertal status and pubertal timing. Psychol Med 28:51-61.

Apter-Levy Y, Feldman M, Vakart A, Ebstein RP, Feldman R (2013) Impact of maternal depression across the first 6 years of life on the child's mental health, social engagement, and empathy: The moderating role of oxytocin. The American journal of psychiatry 170:1161-1168.

Arata CM, Langhinrichsen-Rohling J, Bowers D, O'Brien N (2007) Differential correlates of multi-type maltreatment among urban youth. Child abuse & neglect 31:393-415.

Armony JL, Dolan RJ (2002) Modulation of spatial attention by fear-conditioned stimuli: an event-related fMRI study. Neuropsychologia 40:817-826.

Aupperle RL, Melrose AJ, Francisco A, Paulus MP, Stein MB (2014) Neural substrates of approach-avoidance conflict decision-making. Human brain mapping.

Bakermans-Kranenburg MJ, van IJzendoorn MH (2014) A sociability gene? Meta-analysis of oxytocin receptor genotype effects in humans. Psychiatric Genetics 24:45-51.

Bale TL, Davis AM, Auger AP, Dorsa DM, McCarthy MM (2001) CNS region-specific oxytocin receptor expression: importance in regulation of anxiety and sex behavior. The Journal of neuroscience : the official journal of the Society for Neuroscience 21:2546-2552.

Bartz JA, Hollander E (2006) The neuroscience of affiliation: forging links between basic and clinical research on neuropeptides and social behavior. Hormones and behavior 50:518-528.

Behzadi Y, Restom K, Liau J, Liu TT (2007) A component based noise correction method (CompCor) for BOLD and perfusion based fMRI. NeuroImage 37:90-101.

Bick J, Nelson CA (2016) Early Adverse Experiences and the Developing Brain. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 41:177-196.

Birmaher B, Arbelaez C, Brent D (2002) Course and outcome of child and adolescent major depressive disorder. Child and adolescent psychiatric clinics of North America 11:619-637, x.

Birmaher B, Khetarpal S, Brent D, Cully M, Balach L, Kaufman J, Neer SM (1997) The Screen for Child Anxiety Related Emotional Disorders (SCARED): scale construction and psychometric characteristics. Journal of the American Academy of Child and Adolescent Psychiatry 36:545-553.

Bogdan R, Nikolova YS, Pizzagalli DA (2013) Neurogenetics of depression: a focus on reward processing and stress sensitivity. Neurobiology of disease 52:12-23.

Bogdan R, Pagliaccio D, Baranger DA, Hariri AR (2016) Genetic Moderation of Stress Effects on Corticolimbic Circuitry. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 41:275-296.

Bogdan R, Pizzagalli DA (2006) Acute stress reduces reward responsiveness: implications for depression. Biological psychiatry 60:1147-1154.

Bornstein MH, Arterberry ME (2003) Recognition, discrimination and categorization of smiling by 5-month-old infants. Developmental science 6:585-599.

Botvinick M, Nystrom LE, Fissell K, Carter CS, Cohen JD (1999) Conflict monitoring versus selection-for-action in anterior cingulate cortex. Nature 402:179-181.

Botvinick MM, Braver TS, Barch DM, Carter CS, Cohen JD (2001) Conflict monitoring and cognitive control. Psychological review 108:624-652.

Brawley OW, Freeman HP (1999) Race and outcomes: is this the end of the beginning for minority health research? Journal of the National Cancer Institute 91:1908-1909.

Bronfenbrenner U, Ceci SJ (1994) Nature-nurture reconceptualized in developmental perspective: a bioecological model. Psychological review 101:568-586.

Brown VM, Labar KS, Haswell CC, Gold AL, Mid-Atlantic MW, Beall SK, Van Voorhees E, Marx CE, Calhoun PS, Fairbank JA, Green KT, Tupler LA, Weiner RD, Beckham JC, Brancu M, Hoerle JM, Pender M, Kudler H, Swinkels CM, Nieuwsma JA, Runnals JJ, Youssef NA, McDonald SD, Davison R, Yoash-Gantz R, Taber KH, Hurley R, McCarthy G, Morey RA (2014) Altered resting-state functional connectivity of basolateral and centromedial amygdala complexes in posttraumatic stress disorder. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 39:361-369.

Bruce SE, Buchholz KR, Brown WJ, Yan L, Durbin A, Sheline YI (2012) Altered emotional interference processing in the amygdala and insula in women with Post-Traumatic Stress Disorder. NeuroImage Clinical 2:43-49.

Brune M (2012) Does the oxytocin receptor (OXTR) polymorphism (rs2254298) confer 'vulnerability' for psychopathology or 'differential susceptibility'? Insights from evolution. BMC medicine 10:38.

Buckner RL, Andrews-Hanna JR, Schacter DL (2008) The brain's default network: anatomy, function, and relevance to disease. Annals of the New York Academy of Sciences 1124:1-38.

Burghy CA, Stodola DE, Ruttle PL, Molloy EK, Armstrong JM, Oler JA, Fox ME, Hayes AS, Kalin NH, Essex MJ, Davidson RJ, Birn RM (2012) Developmental pathways to amygdala-prefrontal function and internalizing symptoms in adolescence. Nature neuroscience 15:1736-1741.

Callaghan BL, Tottenham N (2016) The Neuro-Environmental Loop of Plasticity: A Cross-Species Analysis of Parental Effects on Emotion Circuitry Development Following Typical and Adverse Caregiving. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 41:163-176.

Carlson JM, Greenberg T, Rubin D, Mujica-Parodi LR (2011) Feeling anxious: anticipatory amygdalo-insular response predicts the feeling of anxious anticipation. Social cognitive and affective neuroscience 6:74-81.

Carver CS, White TL (1994) Behavioral inhibition, behavioral activation, and affective responses to impending reward and punishment: The BIS/BAS Scales. Journal of Personality and Social Psychology 67:319-333.

Casey BJ, Trainor RJ, Orendi JL, Schubert AB, Nystrom LE, Giedd JN, Castellanos FX, Haxby JV, Noll DC, Cohen JD, Forman SD, Dahl RE, Rapoport JL (1997) A Developmental

Functional MRI Study of Prefrontal Activation during Performance of a Go-No-Go Task. Journal of cognitive neuroscience 9:835-847.

Caspi A, Hariri AR, Holmes A, Uher R, Moffitt TE (2010) Genetic sensitivity to the environment: the case of the serotonin transporter gene and its implications for studying complex diseases and traits. The American journal of psychiatry 167:509-527.

Cauda F, Geminiani GC, Vercelli A (2014) Evolutionary appearance of von Economo's neurons in the mammalian cerebral cortex. Frontiers in human neuroscience 8:104.

Chai XJ, Castanon AN, Ongur D, Whitfield-Gabrieli S (2012) Anticorrelations in resting state networks without global signal regression. NeuroImage 59:1420-1428.

Chen CH, Suckling J, Ooi C, Fu CH, Williams SC, Walsh ND, Mitterschiffthaler MT, Pich EM, Bullmore E (2008) Functional coupling of the amygdala in depressed patients treated with antidepressant medication. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 33:1909-1918.

Chen ZY, Jing D, Bath KG, Ieraci A, Khan T, Siao CJ, Herrera DG, Toth M, Yang C, McEwen BS, Hempstead BL, Lee FS (2006) Genetic variant BDNF (Val66Met) polymorphism alters anxiety-related behavior. Science 314:140-143.

Choe AS, Jones CK, Joel SE, Muschelli J, Belegu V, Caffo BS, Lindquist MA, van Zijl PC, Pekar JJ (2015) Reproducibility and Temporal Structure in Weekly Resting-State fMRI over a Period of 3.5 Years. PloS one 10:e0140134.

Cicchetti D, Rogosch FA Equifinality and multifinality in developmental psychopathology.

Collishaw S, Pickles A, Messer J, Rutter M, Shearer C, Maughan B (2007) Resilience to adult psychopathology following childhood maltreatment: evidence from a community sample. Child abuse & neglect 31:211-229.

Consortium C-DGotPG (2013) Identification of risk loci with shared effects on five major psychiatric disorders: a genome-wide analysis. Lancet (London, England) 381:1371-1379.

Cooney RE, Atlas LY, Joormann J, Eugene F, Gotlib IH (2006) Amygdala activation in the processing of neutral faces in social anxiety disorder: is neutral really neutral? Psychiatry research 148:55-59.

Craig AD (2011) Significance of the insula for the evolution of human awareness of feelings from the body. Annals of the New York Academy of Sciences 1225:72-82.

Critchley HD (2005) Neural mechanisms of autonomic, affective, and cognitive integration. The Journal of comparative neurology 493:154-166.

Crusto CA, Whitson ML, Walling SM, Feinn R, Friedman SR, Reynolds J, Amer M, Kaufman JS (2010) Posttraumatic stress among young urban children exposed to family violence and other potentially traumatic events. Journal of traumatic stress 23:716-724.

Cuthbert BN, Insel TR (2013) Toward the future of psychiatric diagnosis: the seven pillars of RDoC. BMC medicine 11:126.

Dannlowski U, Stuhrmann A, Beutelmann V, Zwanzger P, Lenzen T, Grotegerd D, Domschke K, Hohoff C, Ohrmann P, Bauer J, Lindner C, Postert C, Konrad C, Arolt V, Heindel W, Suslow T, Kugel H (2012) Limbic scars: long-term consequences of childhood maltreatment revealed by functional and structural magnetic resonance imaging. Biological psychiatry 71:286-293.

Darwin C (1897) The Expression of the Emotions in Man and Animals: D. Appleton & Company.

Davis M, Whalen PJ (2001) The amygdala: vigilance and emotion. Molecular psychiatry 6:13-34.

De Bellis MD (2001) Developmental traumatology: the psychobiological development of maltreated children and its implications for research, treatment, and policy. Development and psychopathology 13:539-564.

De Bellis MD, Zisk A (2014) The biological effects of childhood trauma. Child and adolescent psychiatric clinics of North America 23:185-222, vii.

de Moor MHM, van den Berg SM, Verweij KJH, Krueger RF, Luciano M, Vasquez AA, Matteson LK, Derringer J, Esko T, Amin N, Gordon SD, Hansell NK, Hart AB, Seppälä I, Huffman JE, Konte B, Lahti J, Lee M, Miller M, Nutile T, Tanaka T, Teumer A, Viktorin A, Wedenoja J, Abecasis GR, Adkins DE, Agrawal A, Allik J, Appel K, Bigdeli TB, Busonero F, Campbell H, Costa PT, Smith GD, Davies G, de Wit H, Ding J, Engelhardt BE, Eriksson JG, Fedko IO, Ferrucci L, Franke B, Giegling I, Grucza R, Hartmann AM, Heath AC, Heinonen K, Henders AK, Homuth G, Hottenga J-J, Janzing J, Jokela M, Karlsson R, Kemp JP, Kirkpatrick MG, Latvala A, Lehtimäki T, Liewald DC, Madden PAF, Magri C, Magnusson PKE, Marten J, Maschio A, Medland SE, Mihailov E, Milaneschi Y, Montgomery GW, Nauck M, Ouwens KG, Palotie A, Pettersson E, Polasek O, Qian Y, Pulkki-Råback L, Raitakari OT, Realo A, Rose RJ, Ruggiero D, Schmidt CO, Slutske WS, Sorice R, Starr JM, Pourcain BS, Sutin AR, Timpson NJ, Trochet H, Vermeulen S, Vuoksimaa E, Widen E, Wouda J, Wright MJ, Zgaga L, Scotland G, Porteous D, Minelli A, Palmer AA, Rujescu D, Ciullo M, Hayward C, Rudan I, Metspalu A, Kaprio J, Deary IJ, Räikkönen K, Wilson JF, Keltikangas-Järvinen L, Bierut LJ, Hettema JM, Grabe HJ, van Duijn CM, Evans DM, Schlessinger D, Pedersen

NL, Terracciano A, McGue M, Penninx BWJH, Martin NG, Boomsma DI (2015) Genome-wide association study identifies novel locus for neuroticism and shows polygenic association with Major Depressive Disorder. JAMA psychiatry 72:642-650.

Dearing KF, Gotlib IH (2009) Interpretation of ambiguous information in girls at risk for depression. Journal of abnormal child psychology 37:79-91.

Dedovic K, Duchesne A, Andrews J, Engert V, Pruessner JC (2009) The brain and the stress axis: the neural correlates of cortisol regulation in response to stress. NeuroImage 47:864-871.

Deen B, Pelphrey K (2012) Perspective: Brain scans need a rethink. Nature 491:S20.

Derrfuss J, Brass M, Neumann J, von Cramon DY (2005) Involvement of the inferior frontal junction in cognitive control: meta-analyses of switching and Stroop studies. Human brain mapping 25:22-34.

Dillon DG, Holmes AJ, Birk JL, Brooks N, Lyons-Ruth K, Pizzagalli DA (2009) Childhood adversity is associated with left basal ganglia dysfunction during reward anticipation in adulthood. Biological psychiatry 66:206-213.

Divers J, Redden DT, Rice KM, Vaughan LK, Padilla MA, Allison DB, Bluemke DA, Young HJ, Arnett DK (2011) Comparing self-reported ethnicity to genetic background measures in the context of the Multi-Ethnic Study of Atherosclerosis (MESA). BMC genetics 12:28.

Domes G, Heinrichs M, Glascher J, Buchel C, Braus DF, Herpertz SC (2007) Oxytocin attenuates amygdala responses to emotional faces regardless of valence. Biological psychiatry 62:1187-1190.

Domes G, Lischke A, Berger C, Grossmann A, Hauenstein K, Heinrichs M, Herpertz SC (2010) Effects of intranasal oxytocin on emotional face processing in women. Psychoneuroendocrinology 35:83-93.

Drevets WC, Raichle ME (1998) Reciprocal suppression of regional cerebral blood flow during emotional versus higher cognitive processes: Implications for interactions between emotion and cognition. Cognition and Emotion 12:353-385.

Dringenberg HC, Vanderwolf CH (1996) Cholinergic activation of the electrocorticogram: an amygdaloid activating system. Experimental brain research 108:285-296.

Dube SR, Anda RF, Felitti VJ, Chapman DP, Williamson DF, Giles WH (2001) Childhood abuse, household dysfunction, and the risk of attempted suicide throughout the life span: Findings from the adverse childhood experiences study. JAMA 286:3089-3096.

Edwards S, Baynes BB, Carmichael CY, Zamora-Martinez ER, Barrus M, Koob GF, Gilpin NW (2013) Traumatic stress reactivity promotes excessive alcohol drinking and alters the balance of prefrontal cortex-amygdala activity. Translational psychiatry 3:e296.

Egger HL, Pine DS, Nelson E, Leibenluft E, Ernst M, Towbin KE, Angold A (2011) The NIMH Child Emotional Faces Picture Set (NIMH-ChEFS): a new set of children's facial emotion stimuli. International journal of methods in psychiatric research 20:145-156.

Egner T, Etkin A, Gale S, Hirsch J (2008) Dissociable neural systems resolve conflict from emotional versus nonemotional distracters. Cerebral cortex 18:1475-1484.

Eickhoff SB, Stephan KE, Mohlberg H, Grefkes C, Fink GR, Amunts K, Zilles K (2005) A new SPM toolbox for combining probabilistic cytoarchitectonic maps and functional imaging data. NeuroImage 25:1325-1335.

Ekman P (1992) An argument for basic emotions. Cognition & emotion 6:169-200.

Ekman P, Friesen WV (1976) Pictures of Facial Affect: Consulting psychologists Press.

English DJ, Upadhyaya MP, Litrownik AJ, Marshall JM, Runyan DK, Graham JC, Dubowitz H (2005) Maltreatment's wake: the relationship of maltreatment dimensions to child outcomes. Child abuse & neglect 29:597-619.

Etkin A, Egner T, Kalisch R (2011) Emotional processing in anterior cingulate and medial prefrontal cortex. Trends in cognitive sciences 15:85-93.

Etkin A, Egner T, Peraza DM, Kandel ER, Hirsch J (2006) Resolving emotional conflict: a role for the rostral anterior cingulate cortex in modulating activity in the amygdala. Neuron 51:871-882.

Etkin A, Gyurak A, O'Hara R (2013) A neurobiological approach to the cognitive deficits of psychiatric disorders. Dialogues in clinical neuroscience 15:419-429.

Etkin A, Prater KE, Hoeft F, Menon V, Schatzberg AF (2010) Failure of anterior cingulate activation and connectivity with the amygdala during implicit regulation of emotional processing in generalized anxiety disorder. The American journal of psychiatry 167:545-554.

Etkin A, Prater KE, Schatzberg AF, Menon V, Greicius MD (2009) Disrupted amygdalar subregion functional connectivity and evidence of a compensatory network in generalized anxiety disorder. Archives of general psychiatry 66:1361-1372.

Etkin A, Schatzberg AF (2011) Common abnormalities and disorder-specific compensation during implicit regulation of emotional processing in generalized anxiety and major depressive disorders. The American journal of psychiatry 168:968-978.

Etkin A, Wager TD (2007) Functional neuroimaging of anxiety: a meta-analysis of emotional processing in PTSD, social anxiety disorder, and specific phobia. The American journal of psychiatry 164:1476-1488.

Fair DA, Nigg JT, Iyer S, Bathula D, Mills KL, Dosenbach NU, Schlaggar BL, Mennes M, Gutman D, Bangaru S, Buitelaar JK, Dickstein DP, Di Martino A, Kennedy DN, Kelly C, Luna B, Schweitzer JB, Velanova K, Wang YF, Mostofsky S, Castellanos FX, Milham MP (2012) Distinct neural signatures detected for ADHD subtypes after controlling for micro-movements in resting state functional connectivity MRI data. Frontiers in systems neuroscience 6:80.

Falk EB, Hyde LW, Mitchell C, Faul J, Gonzalez R, Heitzeg MM, Keating DP, Langa KM, Martz ME, Maslowsky J, Morrison FJ, Noll DC, Patrick ME, Pfeffer FT, Reuter-Lorenz PA, Thomason ME, Davis-Kean P, Monk CS, Schulenberg J (2013) What is a representative brain? Neuroscience meets population science. Proceedings of the National Academy of Sciences of the United States of America 110:17615-17622.

Felitti VJ, Anda RF, Nordenberg D, Williamson DF, Spitz AM, Edwards V, Koss MP, Marks JS (1998) Relationship of childhood abuse and household dysfunction to many of the leading causes of death in adults. The Adverse Childhood Experiences (ACE) Study. American journal of preventive medicine 14:245-258.

Ferrari AJ, Charlson FJ, Norman RE, Patten SB, Freedman G, Murray CJ, Vos T, Whiteford HA (2013) Burden of depressive disorders by country, sex, age, and year: findings from the global burden of disease study 2010. PLoS medicine 10:e1001547.

Finkelhor D, Turner HA, Shattuck A, Hamby SL (2013) Violence, crime, and abuse exposure in a national sample of children and youth: an update. JAMA pediatrics 167:614-621.

Foland-Ross LC, Gotlib IH (2012) Cognitive and neural aspects of information processing in major depressive disorder: an integrative perspective. Frontiers in psychology 3:489.

Foland-Ross LC, Hardin MG, Gotlib IH (2013) Neurobiological markers of familial risk for depression. Current topics in behavioral neurosciences 14:181-206.

Forbes EE, Dahl RE (2005) Neural systems of positive affect: relevance to understanding child and adolescent depression? Development and psychopathology 17:827-850.

Forbes EE, Dahl RE (2010) Pubertal development and behavior: hormonal activation of social and motivational tendencies. Brain and cognition 72:66-72.

Forbes EE, Hariri AR, Martin SL, Silk JS, Moyles DL, Fisher PM, Brown SM, Ryan ND, Birmaher B, Axelson DA, Dahl RE (2009) Altered striatal activation predicting real-world positive affect in adolescent major depressive disorder. The American journal of psychiatry 166:64-73.

Forbes EE, Phillips ML, Silk JS, Ryan ND, Dahl RE (2011) Neural systems of threat processing in adolescents: role of pubertal maturation and relation to measures of negative affect. Developmental neuropsychology 36:429-452.

Forbes EE, Ryan ND, Phillips ML, Manuck SB, Worthman CM, Moyles DL, Tarr JA, Sciarrillo SR, Dahl RE (2010) Healthy adolescents' neural response to reward: associations with puberty, positive affect, and depressive symptoms. Journal of the American Academy of Child and Adolescent Psychiatry 49:162-172.e161-165.

Ford JDP (2005) Treatment Implications of Altered Affect Regulation and Information Processing Following Child Maltreatment. Psychiatric Annals 35:410-419.

Fox MD, Snyder AZ, Vincent JL, Corbetta M, Van Essen DC, Raichle ME (2005) The human brain is intrinsically organized into dynamic, anticorrelated functional networks.

Proceedings of the National Academy of Sciences of the United States of America 102:9673-9678.

Fransson P, Aden U, Blennow M, Lagercrantz H (2011) The functional architecture of the infant brain as revealed by resting-state fMRI. Cerebral cortex 21:145-154.

Friston KJ, Buechel C, Fink GR, Morris J, Rolls E, Dolan RJ (1997) Psychophysiological and modulatory interactions in neuroimaging. NeuroImage 6:218-229.

Furman DJ, Chen MC, Gotlib IH (2011) Variant in oxytocin receptor gene is associated with amygdala volume. Psychoneuroendocrinology 36:891-897.

Fusar-Poli P, Howes O, Bechdolf A, Borgwardt S (2012) Mapping vulnerability to bipolar disorder: a systematic review and meta-analysis of neuroimaging studies. Journal of psychiatry & neuroscience : JPN 37:170-184.

Fusar-Poli P, Placentino A, Carletti F, Landi P, Allen P, Surguladze S, Benedetti F, Abbamonte M, Gasparotti R, Barale F, Perez J, McGuire P, Politi P (2009) Functional atlas of emotional faces processing: a voxel-based meta-analysis of 105 functional magnetic resonance imaging studies. Journal of psychiatry & neuroscience : JPN 34:418-432.

Gabard-Durnam LJ, Flannery J, Goff B, Gee DG, Humphreys KL, Telzer E, Hare T, Tottenham N (2014) The development of human amygdala functional connectivity at rest from 4 to 23 years: a cross-sectional study. NeuroImage 95:193-207.

Gabbay V, Johnson AR, Alonso CM, Evans LK, Babb JS, Klein RG (2015) Anhedonia, but not irritability, is associated with illness severity outcomes in adolescent major depression. Journal of child and adolescent psychopharmacology 25:194-200.

Gambarana C, Masi F, Tagliamonte A, Scheggi S, Ghiglieri O, De Montis MG (1999) A chronic stress that impairs reactivity in rats also decreases dopaminergic transmission in the nucleus accumbens: a microdialysis study. J Neurochem 72:2039-2046.

Gamer M, Zurowski B, Büchel C (2010) Different amygdala subregions mediate valence-related and attentional effects of oxytocin in humans. Proceedings of the National Academy of Sciences of the United States of America 107:9400-9405.

Gao W, Gilmore JH, Giovanello KS, Smith JK, Shen D, Zhu H, Lin W (2011) Temporal and spatial evolution of brain network topology during the first two years of life. PloS one 6:e25278.

Garner KG, Dux PE, Wagner J, Cummins TD, Chambers CD, Bellgrove MA (2012) Attentional asymmetries in a visual orienting task are related to temperament. Cognition & emotion 26:1508-1515.

Gee DG, Gabard-Durnam LJ, Flannery J, Goff B, Humphreys KL, Telzer EH, Hare TA, Bookheimer SY, Tottenham N (2013a) Early developmental emergence of human amygdala-prefrontal connectivity after maternal deprivation. Proceedings of the National Academy of Sciences of the United States of America 110:15638-15643.

Gee DG, Humphreys KL, Flannery J, Goff B, Telzer EH, Shapiro M, Hare TA, Bookheimer SY, Tottenham N (2013b) A developmental shift from positive to negative connectivity in human amygdala-prefrontal circuitry. The Journal of neuroscience : the official journal of the Society for Neuroscience 33:4584-4593.

Giedd JN, Keshavan M, Paus T (2008) Why do many psychiatric disorders emerge during adolescence? Nature reviews Neuroscience 9:947-957.

Gilbert R, Widom CS, Browne K, Fergusson D, Webb E, Janson S (2009) Burden and consequences of child maltreatment in high-income countries. Lancet 373:68-81.

Gillespie CF, Bradley B, Mercer K, Smith AK, Conneely K, Gapen M, Weiss T, Schwartz AC, Cubells JF, Ressler KJ (2009) Trauma exposure and stress-related disorders in inner city primary care patients. General hospital psychiatry 31:505-514.

Glaser D (2000) Child abuse and neglect and the brain--a review. Journal of child psychology and psychiatry, and allied disciplines 41:97-116.

Glowinski AL, Madden PA, Bucholz KK, Lynskey MT, Heath AC (2003) Genetic epidemiology of self-reported lifetime DSM-IV major depressive disorder in a population-based twin sample of female adolescents. Journal of child psychology and psychiatry, and allied disciplines 44:988-996.

Gobbini MI, Haxby JV (2007) Neural systems for recognition of familiar faces. Neuropsychologia 45:32-41.

Godlewska BR, Norbury R, Selvaraj S, Cowen PJ, Harmer CJ (2012) Short-term SSRI treatment normalises amygdala hyperactivity in depressed patients. Psychological medicine 42:2609-2617.

Goforth A, Pham A, Carlson J (2011) Diathesis-stress Model. In: Encyclopedia of Child Behavior and Development (Goldstein, S. and Naglieri, J., eds), pp 502-503: Springer US.

Gogtay N, Giedd JN, Lusk L, Hayashi KM, Greenstein D, Vaituzis AC, Nugent TF, 3rd, Herman DH, Clasen LS, Toga AW, Rapoport JL, Thompson PM (2004) Dynamic mapping of human cortical development during childhood through early adulthood. Proceedings of the National Academy of Sciences of the United States of America 101:8174-8179.

Goldmann E, Aiello A, Uddin M, Delva J, Koenen K, Gant LM, Galea S (2011) Pervasive exposure to violence and posttraumatic stress disorder in a predominantly African American Urban Community: the Detroit Neighborhood Health Study. Journal of traumatic stress 24:747-751.

Goossens L, Kukolja J, Onur OA, Fink GR, Maier W, Griez E, Schruers K, Hurlemann R (2009) Selective processing of social stimuli in the superficial amygdala. Human brain mapping 30:3332-3338.

Grant MM, White D, Hadley J, Hutcheson N, Shelton R, Sreenivasan K, Deshpande G (2014) Early life trauma and directional brain connectivity within major depression. Human brain mapping.

Green JG, McLaughlin KA, Berglund PA, Gruber MJ, Sampson NA, Zaslavsky AM, Kessler RC (2010) Childhood adversities and adult psychiatric disorders in the national comorbidity survey replication I: associations with first onset of DSM-IV disorders. Archives of general psychiatry 67:113-123.

Greenberg PE, Kessler RC, Birnbaum HG, Leong SA, Lowe SW, Berglund PA, Corey-Lisle PK (2003) The economic burden of depression in the United States: how did it change between 1990 and 2000? The Journal of clinical psychiatry 64:1465-1475.

Greicius M (2008) Resting-state functional connectivity in neuropsychiatric disorders. Current opinion in neurology 21:424-430.

Greicius MD, Supekar K, Menon V, Dougherty RF (2009) Resting-state functional connectivity reflects structural connectivity in the default mode network. Cerebral cortex 19:72-78.

Gyurak A, Gross JJ, Etkin A (2011) Explicit and implicit emotion regulation: a dual-process framework. Cognition & emotion 25:400-412.

Haber SN, Knutson B (2010) The reward circuit: linking primate anatomy and human imaging. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 35:4-26.

Hakamata Y, Lissek S, Bar-Haim Y, Britton JC, Fox NA, Leibenluft E, Ernst M, Pine DS (2010) Attention bias modification treatment: a meta-analysis toward the establishment of novel treatment for anxiety. Biological psychiatry 68:982-990.

Hamilton JP, Chen G, Thomason ME, Schwartz ME, Gotlib IH (2011) Investigating neural primacy in Major Depressive Disorder: multivariate Granger causality analysis of resting-state fMRI time-series data. Molecular psychiatry 16:763-772.

Hamilton JP, Etkin A, Furman DJ, Lemus MG, Johnson RF, Gotlib IH (2012) Functional neuroimaging of major depressive disorder: a meta-analysis and new integration of base line activation and neural response data. The American journal of psychiatry 169:693-703.

Hanson JL, Albert D, Iselin AR, Carre JM, Dodge KA, Hariri AR (2015) Cumulative stress in childhood is associated with blunted reward-related brain activity in adulthood. Social cognitive and affective neuroscience.

Hardy JL, Nelson RA, Thomason ME, Sternberg DA, Katovich K, Farzin F, Scanlon M (2015) Enhancing Cognitive Abilities with Comprehensive Training: A Large, Online, Randomized, Active-Controlled Trial. PloS one 10:e0134467.

Hare TA, Tottenham N, Galvan A, Voss HU, Glover GH, Casey BJ (2008) Biological substrates of emotional reactivity and regulation in adolescence during an emotional go-nogo task. Biological psychiatry 63:927-934.

Hariri AR, Mattay VS, Tessitore A, Kolachana B, Fera F, Goldman D, Egan MF, Weinberger DR (2002a) Serotonin transporter genetic variation and the response of the human amygdala. Science 297:400-403.

Hariri AR, Tessitore A, Mattay VS, Fera F, Weinberger DR (2002b) The Amygdala Response to Emotional Stimuli: A Comparison of Faces and Scenes. NeuroImage 17:317-323.

Hartley CA, Lee FS (2015) Sensitive periods in affective development: nonlinear maturation of fear learning. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 40:50-60.

Hasler G, Drevets WC, Manji HK, Charney DS (2004) Discovering endophenotypes for major depression. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 29:1765-1781.

Hayes AF (2013) Introduction to Mediation, Moderation, and Conditional Process Analysis: A Regression-Based Approach: Guilford Publications.

Heinrichs M, Baumgartner T, Kirschbaum C, Ehlert U (2003) Social support and oxytocin interact to suppress cortisol and subjective responses to psychosocial stress. Biological psychiatry 54:1389-1398.

Herba C, Phillips M (2004) Annotation: Development of facial expression recognition from childhood to adolescence: behavioural and neurological perspectives. Journal of Child Psychology and Psychiatry 45:1185-1198.

Herringa RJ, Birn RM, Ruttle PL, Burghy CA, Stodola DE, Davidson RJ, Essex MJ (2013a) Childhood maltreatment is associated with altered fear circuitry and increased internalizing symptoms by late adolescence. Proceedings of the National Academy of Sciences of the United States of America 110:19119-19124.

Herringa RJ, Phillips ML, Fournier JC, Kronhaus DM, Germain A (2013b) Childhood and adult trauma both correlate with dorsal anterior cingulate activation to threat in combat veterans. Psychological medicine 43:1533-1542.

Hickey C, Chelazzi L, Theeuwes J (2010) Reward guides vision when it's your thing: trait reward-seeking in reward-mediated visual priming. PloS one 5:e14087.

Holmes AJ, Lee PH, Hollinshead MO, Bakst L, Roffman JL, Smoller JW, Buckner RL (2012) Individual differences in amygdala-medial prefrontal anatomy link negative affect, impaired social functioning, and polygenic depression risk. The Journal of neuroscience : the official journal of the Society for Neuroscience 32:18087-18100.

Holz NE, Buchmann AF, Boecker R, Blomeyer D, Baumeister S, Wolf I, Rietschel M, Witt SH, Plichta MM, Meyer-Lindenberg A, Banaschewski T, Brandeis D, Laucht M (2015) Role of FKBP5 in emotion processing: results on amygdala activity, connectivity and volume. Brain structure & function 220:1355-1368.

Hubel DH, Wiesel TN (2004) Brain and Visual Perception: The Story of a 25-Year Collaboration. Oxford: Oxford University Press.

Huttenlocher PR, de Courten C, Garey LJ, Van der Loos H (1982) Synaptogenesis in human visual cortex--evidence for synapse elimination during normal development. Neuroscience letters 33:247-252.

Inglis FM, Moghaddam B (1999) Dopaminergic innervation of the amygdala is highly responsive to stress. J Neurochem 72:1088-1094.

Inoue H, Yamasue H, Tochigi M, Abe O, Liu X, Kawamura Y, Takei K, Suga M, Yamada H, Rogers MA, Aoki S, Sasaki T, Kasai K (2010) Association between the oxytocin receptor gene and amygdalar volume in healthy adults. Biological psychiatry 68:1066-1072.

Ippen CG, Ford J, Racusin R, Acker M, K. B, Rogers C, Ellis C, Schiffman J, Ribbe D, Cone P, Lukovitz M, Edwards J (2002) Traumatic Events Screening Inventory - Parent Report Revised. San Francisco, CA: Early Trauma Network and the National Center for PTSD Dartmouth Child Trauma Research Group.

Jacobson NS, Martell CR, Dimidjian S (2001) Behavioral Activation Treatment for Depression: Returning to Contextual Roots. Clinical Psychology: Science and Practice 8:255-270.

Ji J, Maren S (2007) Hippocampal involvement in contextual modulation of fear extinction. Hippocampus 17:749-758.

Joiner TE, Jr., Lewinsohn PM, Seeley JR (2002) The core of loneliness: lack of pleasurable engagement--more so than painful disconnection--predicts social impairment, depression onset, and recovery from depressive disorders among adolescents. Journal of personality assessment 79:472-491.

Joshi A, Leahy RM, Toga A, Shattuck DW (2009) A Framework for Brain Registration via Simultaneous Surface and Volume Flow. Information processing in medical imaging : proceedings of the conference 21:576-588.

Joshi A, Shattuck DW, Thompson PM, Leahy RM (2007) Surface-constrained volumetric brain registration using harmonic mappings. IEEE transactions on medical imaging 26:1657-1669.

Kalisch R, Wiech K, Critchley HD, Seymour B, O'Doherty JP, Oakley DA, Allen P, Dolan RJ (2005) Anxiety reduction through detachment: subjective, physiological, and neural effects. Journal of cognitive neuroscience 17:874-883.

Kalueff AV, Olivier JDA, Nonkes LJP, Homberg JR (2010) Conserved role for the serotonin transporter gene in rat and mouse neurobehavioral endophenotypes. Neuroscience & Biobehavioral Reviews 34:373-386.

Kasch KL, Rottenberg J, Arnow BA, Gotlib IH (2002) Behavioral activation and inhibition systems and the severity and course of depression. Journal of Abnormal Psychology 111:589-597.

Kashdan TB, Elhai JD, Frueh BC (2006) Anhedonia and emotional numbing in combat veterans with PTSD. Behaviour research and therapy 44:457-467.

Kaufman AS, Kaufman NL (2004a) Kaufman Brief Intelligence Test-Second Edition (KBIT-2): Circle Pines, MN: American Guidance Service. .

Kaufman AS, Kaufman NL (2004b) Kaufman Brief Intelligence Test: KBIT 2 ; Manual: Pearson.

Kaufman J, Charney D (2000) Comorbidity of mood and anxiety disorders. Depression and anxiety 12 Suppl 1:69-76.

Kaufman J, Plotsky PM, Nemeroff CB, Charney DS (2000) Effects of early adverse experiences on brain structure and function: clinical implications. Biological psychiatry 48:778-790.

Kennard B, Silva S, Vitiello B, Curry J, Kratochvil C, Simons A, Hughes J, Feeny N, Weller E, Sweeney M, Reinecke M, Pathak S, Ginsburg G, Emslie G, March J (2006) Remission and residual symptoms after short-term treatment in the Treatment of Adolescents with Depression Study (TADS). Journal of the American Academy of Child and Adolescent Psychiatry 45:1404-1411.

Kessler RC, Akiskal HS, Ames M, Birnbaum H, Greenberg P, Hirschfeld RM, Jin R, Merikangas KR, Simon GE, Wang PS (2006) Prevalence and effects of mood disorders

on work performance in a nationally representative sample of U.S. workers. The American journal of psychiatry 163:1561-1568.

Kessler RC, Avenevoli S, Ries Merikangas K (2001) Mood disorders in children and adolescents: an epidemiologic perspective. Biological psychiatry 49:1002-1014.

Kessler RC, Berglund P, Demler O, Jin R, Merikangas KR, Walters EE (2005a) Lifetime prevalence and age-of-onset distributions of DSM-IV disorders in the National Comorbidity Survey Replication. Archives of general psychiatry 62:593-602.

Kessler RC, Chiu WT, Demler O, Merikangas KR, Walters EE (2005b) Prevalence, severity, and comorbidity of 12-month DSM-IV disorders in the National Comorbidity Survey Replication. Archives of general psychiatry 62:617-627.

Kessler RC, Davis CG, Kendler KS (1997) Childhood adversity and adult psychiatric disorder in the US National Comorbidity Survey. Psychological Medicine 27:1101-1119.

Kessler RC, McLaughlin KA, Green JG, Gruber MJ, Sampson NA, Zaslavsky AM, Aguilar-Gaxiola S, Alhamzawi AO, Alonso J, Angermeyer M, Benjet C, Bromet E, Chatterji S, de Girolamo G, Demyttenaere K, Fayyad J, Florescu S, Gal G, Gureje O, Haro JM, Hu CY, Karam EG, Kawakami N, Lee S, Lepine JP, Ormel J, Posada-Villa J, Sagar R, Tsang A, Ustun TB, Vassilev S, Viana MC, Williams DR (2010) Childhood adversities and adult psychopathology in the WHO World Mental Health Surveys. The British journal of psychiatry : the journal of mental science 197:378-385.

Kessler RC, Sonnega A, Bromet E, Hughes M, Nelson CB (1995) Posttraumatic stress disorder in the National Comorbidity Survey. Archives of general psychiatry 52:1048-1060.

Kim JE, Lyoo I, Estes AM, et al. (2010) Laterobasal amygdalar enlargement in 6- to 7-year-old children with autism spectrum disorder. Archives of general psychiatry 67:1187-1197.

Kim MJ, Whalen PJ (2009) The structural integrity of an amygdala-prefrontal pathway predicts trait anxiety. The Journal of neuroscience : the official journal of the Society for Neuroscience 29:11614-11618.

Kirsch P, Esslinger C, Chen Q, Mier D, Lis S, Siddhanti S, Gruppe H, Mattay VS, Gallhofer B, Meyer-Lindenberg A (2005) Oxytocin modulates neural circuitry for social cognition and fear in humans. The Journal of neuroscience : the official journal of the Society for Neuroscience 25:11489-11493.

Klüver H, Bucy PC (1937) " Psychic blindness" and other symptoms following bilateral temporal lobectomy in Rhesus monkeys. American Journal of Physiology.

Kosfeld M, Heinrichs M, Zak PJ, Fischbacher U, Fehr E (2005) Oxytocin increases trust in humans. Nature 435:673-676.

Kovacs M (1985) The Children's Depression, Inventory (CDI). Psychopharmacology bulletin 21:995-998.

Kovacs M (1992) Children's Depression Inventory: Multi-Health Systems, Incorporated.

Kraemer HC, Yesavage JA, Taylor JL, Kupfer D (2000) How can we learn about developmental processes from cross-sectional studies, or can we? The American journal of psychiatry 157:163-171.

Lantz PM, House JS, Mero RP, Williams DR (2005) Stress, life events, and socioeconomic disparities in health: results from the Americans' Changing Lives Study. Journal of health and social behavior 46:274-288.

LeDoux J (1998) Fear and the brain: where have we been, and where are we going? Biological psychiatry 44:1229-1238.

LeDoux J (2003) The emotional brain, fear, and the amygdala. Cellular and molecular neurobiology 23:727-738.

LeDoux JE (1996) The emotional brain: the mysterious underpinnings of emotional life: Simon & Schuster.

LeDoux JE (2000) Emotion circuits in the brain. Annual review of neuroscience 23:155-184.

Levran O, Awolesi O, Shen PH, Adelson M, Kreek MJ (2012) Estimating ancestral proportions in a multi-ethnic US sample: implications for studies of admixed populations. Human genomics 6:2.

Levy-Gigi E, Kelemen O, Gluck MA, Keri S (2011) Impaired context reversal learning, but not cue reversal learning, in patients with amnestic mild cognitive impairment. Neuropsychologia 49:3320-3326.

Levy-Gigi E, Richter-Levin G (2014) The hidden price of repeated traumatic exposure. Stress (Amsterdam, Netherlands) 17:343-351.

Lewinsohn PM, Clarke GN, Seeley JR, Rohde P (1994) Major depression in community adolescents: age at onset, episode duration, and time to recurrence. Journal of the American Academy of Child and Adolescent Psychiatry 33:809-818.

Lewinsohn PM, Rohde P, Klein DN, Seeley JR (1999) Natural course of adolescent major depressive disorder: I. Continuity into young adulthood. Journal of the American Academy of Child and Adolescent Psychiatry 38:56-63.

Lieberman MD, Cunningham WA (2009) Type I and Type II error concerns in fMRI research: re-balancing the scale. Social cognitive and affective neuroscience 4:423-428.

Liston C, Chen AC, Zebley BD, Drysdale AT, Gordon R, Leuchter B, Voss HU, Casey BJ, Etkin A, Dubin MJ (2014) Default mode network mechanisms of transcranial magnetic stimulation in depression. Biological psychiatry 76:517-526.

Lobaugh NJ, Gibson E, Taylor MJ (2006) Children recruit distinct neural systems for implicit emotional face processing. Neuroreport 17:215-219.

Lowe SR, Galea S, Uddin M, Koenen KC (2014) Trajectories of posttraumatic stress among urban residents. American journal of community psychology 53:159-172.

Lukas M, Toth I, Reber SO, Slattery DA, Veenema AH, Neumann ID (2011) The neuropeptide oxytocin facilitates pro-social behavior and prevents social avoidance in rats and mice. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 36:2159-2168.

Lutz A, McFarlin DR, Perlman DM, Salomons TV, Davidson RJ (2013) Altered anterior insula activation during anticipation and experience of painful stimuli in expert meditators. NeuroImage 64:538-546.

MacLean PD (1949) Psychosomatic disease and the visceral brain; recent developments bearing on the Papez theory of emotion. Psychosomatic medicine 11:338-353.

Mahan AL, Ressler KJ (2012) Fear conditioning, synaptic plasticity and the amygdala: implications for posttraumatic stress disorder. Trends in neurosciences 35:24-35.

Maheu FS, Dozier M, Guyer AE, Mandell D, Peloso E, Poeth K, Jenness J, Lau JY, Ackerman JP, Pine DS, Ernst M (2010) A preliminary study of medial temporal lobe function in youths with a history of caregiver deprivation and emotional neglect. Cognitive, affective & behavioral neuroscience 10:34-49.

Maier ME, di Pellegrino G (2012) Impaired conflict adaptation in an emotional task context following rostral anterior cingulate cortex lesions in humans. Journal of cognitive neuroscience 24:2070-2079.

Malter Cohen M, Jing D, Yang RR, Tottenham N, Lee FS, Casey BJ (2013a) Early-life stress has persistent effects on amygdala function and development in mice and humans. Proceedings of the National Academy of Sciences of the United States of America 110:18274-18278.

Malter Cohen M, Tottenham N, Casey BJ (2013b) Translational developmental studies of stress on brain and behavior: implications for adolescent mental health and illness? Neuroscience 249:53-62.

Manoliu A, Meng C, Brandl F, Doll A, Tahmasian M, Scherr M, Schwerthoffer D, Zimmer C, Forstl H, Bauml J, Riedl V, Wohlschlager AM, Sorg C (2013) Insular dysfunction within the salience network is associated with severity of symptoms and aberrant inter-network connectivity in major depressive disorder. Frontiers in human neuroscience 7:930.

Marchand WR (2014) Neural mechanisms of mindfulness and meditation: Evidence from neuroimaging studies. World journal of radiology 6:471-479.

Marshall AD (2016) Developmental Timing of Trauma Exposure Relative to Puberty and the Nature of Psychopathology Among Adolescent Girls. Journal of the American Academy of Child and Adolescent Psychiatry 55:25-32 e21.

Marshall WA, Tanner JM (1968) Growth and physiological development during adolescence. Annual review of medicine 19:283-300.

Marusak HA, Carre JM, Thomason ME (2013) The stimuli drive the response: an fMRI study of youth processing adult or child emotional face stimuli. NeuroImage 83:679-689.

Marusak HA, Etkin A, Thomason ME (2015a) Disrupted insula-based connectome organization and conflict interference in trauma-exposed youth. NeuroImage: Clinical In Press.

Marusak HA, Kuruvadi N, Vila AM, Shattuck DW, Joshi SH, Joshi AA, Jella PK, Thomason ME (2015b) Interactive effects of BDNF Val66Met genotype and trauma on limbic brain anatomy in childhood. European child & adolescent psychiatry.

Marusak HA, Martin KA, Etkin A, Thomason ME (2014a) Childhood trauma exposure disrupts the automatic regulation of emotional processing. Neuropsychophamarcology In Press.

Marusak HA, Martin KR, Etkin A, Thomason ME (2014b) Childhood Trauma Exposure Disrupts the Automatic Regulation of Emotional Processing. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology.

McClure EB, Monk CS, Nelson EE, Parrish JM, Adler A, Blair RJ, Fromm S, Charney DS, Leibenluft E, Ernst M, Pine DS (2007) Abnormal attention modulation of fear circuit function in pediatric generalized anxiety disorder. Archives of general psychiatry 64:97-106.

McCrory EJ, De Brito SA, Kelly PA, Bird G, Sebastian CL, Mechelli A, Samuel S, Viding E (2013) Amygdala activation in maltreated children during pre-attentive emotional processing. The British journal of psychiatry : the journal of mental science 202:269-276.

McCrory EJ, De Brito SA, Sebastian CL, Mechelli A, Bird G, Kelly PA, Viding E (2011) Heightened neural reactivity to threat in child victims of family violence. Current biology : CB 21:R947-948.

McDonald AJ (1998) Cortical pathways to the mammalian amygdala. Progress in Neurobiology 55:257-332.

McEwen BS (2012) Brain on stress: how the social environment gets under the skin. Proceedings of the National Academy of Sciences of the United States of America 109 Suppl 2:17180-17185.

McEwen BS, Morrison JH (2013) The brain on stress: vulnerability and plasticity of the prefrontal cortex over the life course. Neuron 79:16-29.

McGee AW, Yang Y, Fischer QS, Daw NW, Strittmatter SM (2005) Experience-driven plasticity of visual cortex limited by myelin and Nogo receptor. Science (New York, NY) 309:2222-2226.

McKenna F (1986) Effects of unattended emotional stimuli on color-naming performance. Current Psychology 5:3-9.

McLaughlin KA, Greif Green J, Gruber MJ, Sampson NA, Zaslavsky AM, Kessler RC (2012) Childhood adversities and first onset of psychiatric disorders in a national sample of US adolescents. Archives of general psychiatry 69:1151-1160.

McLaughlin KA, Sheridan MA, Lambert HK (2014) Childhood adversity and neural development: deprivation and threat as distinct dimensions of early experience. Neuroscience and biobehavioral reviews 47:578-591.

McMakin DL, Olino TM, Porta G, Dietz LJ, Emslie G, Clarke G, Wagner KD, Asarnow JR, Ryan ND, Birmaher B, Shamseddeen W, Mayes T, Kennard B, Spirito A, Keller M, Lynch FL, Dickerson JF, Brent DA (2012) Anhedonia predicts poorer recovery among youth with selective serotonin reuptake inhibitor treatment-resistant depression. Journal of the American Academy of Child and Adolescent Psychiatry 51:404-411.

McMakin DL, Siegle GJ, Shirk SR (2011) Positive Affect Stimulation and Sustainment (PASS) Module for Depressed Mood: A preliminary investigation of treatment-related effects. Cognitive therapy and research 35:217-226.

Mehta MA, Golembo NI, Nosarti C, Colvert E, Mota A, Williams SC, Rutter M, Sonuga-Barke EJ (2009) Amygdala, hippocampal and corpus callosum size following severe early institutional deprivation: the English and Romanian Adoptees study pilot. Journal of child psychology and psychiatry, and allied disciplines 50:943-951.

Meinlschmidt G, Heim C (2007) Sensitivity to intranasal oxytocin in adult men with early parental separation. Biological psychiatry 61:1109-1111.

Menon V (2011) Large-scale brain networks and psychopathology: a unifying triple network model. Trends in cognitive sciences 15:483-506.

Menon V (2013) Developmental pathways to functional brain networks: emerging principles. Trends in cognitive sciences 17:627-640.

Menon V, Uddin LQ (2010) Saliency, switching, attention and control: a network model of insula function. Brain structure & function 214:655-667.

Merboldt KD, Fransson P, Bruhn H, Frahm J (2001) Functional MRI of the human amygdala? NeuroImage 14:253-257.

Mesman J, van Ijzendoorn MH, Bakermans-Kranenburg MJ (2009) The many faces of the Still-Face Paradigm: A review and meta-analysis. Developmental Review 29:120-162.

Mesulam MM, Mufson EJ (1982) Insula of the old world monkey. III: Efferent cortical output and comments on function. The Journal of comparative neurology 212:38-52.

Meyer-Lindenberg A, Weinberger DR (2006) Intermediate phenotypes and genetic mechanisms of psychiatric disorders. Nature reviews Neuroscience 7:818-827.

Milad MR, Pitman RK, Ellis CB, Gold AL, Shin LM, Lasko NB, Zeidan MA, Handwerger K, Orr SP, Rauch SL (2009) Neurobiological basis of failure to recall extinction memory in posttraumatic stress disorder. Biological psychiatry 66:1075-1082.

Morgan JK, Olino TM, McMakin DL, Ryan ND, Forbes EE (2013) Neural response to reward as a predictor of increases in depressive symptoms in adolescence. Neurobiology of disease 52:66-74.

Moriceau S, Shionoya K, Jakubs K, Sullivan RM (2009) Early-life stress disrupts attachment learning: the role of amygdala corticosterone, locus ceruleus corticotropin releasing hormone, and olfactory bulb norepinephrine. The Journal of neuroscience : the official journal of the Society for Neuroscience 29:15745-15755.

Morris SE, Cuthbert BN (2012) Research Domain Criteria: cognitive systems, neural circuits, and dimensions of behavior. Dialogues in clinical neuroscience 14:29-37.

Mosconi MW, Cody-Hazlett H, Poe MD, Gerig G, Gimpel-Smith R, Piven J (2009) Longitudinal study of amygdala volume and joint attention in 2- to 4-year-old children with autism. Archives of general psychiatry 66:509-516.

Mottolese R, Redoute J, Costes N, Le Bars D, Sirigu A (2014) Switching brain serotonin with oxytocin. Proceedings of the National Academy of Sciences of the United States of America 111:8637-8642.

Mukherjee P, Whalley HC, McKirdy JW, McIntosh AM, Johnstone EC, Lawrie SM, Hall J (2011) Effects of the BDNF Val66Met polymorphism on neural responses to facial emotion. Psychiatry research 191:182-188.

Nabel EM, Morishita H (2013) Regulating critical period plasticity: insight from the visual system to fear circuitry for therapeutic interventions. Frontiers in psychiatry 4:146.

Narr KL, Woods RP, Thompson PM, Szeszko P, Robinson D, Dimtcheva T, Gurbani M, Toga AW, Bilder RM (2007) Relationships between IQ and regional cortical gray matter thickness in healthy adults. Cerebral cortex 17:2163-2171.

Nikolova YS, Bogdan R, Brigidi BD, Hariri AR (2012) Ventral striatum reactivity to reward and recent life stress interact to predict positive affect. Biological psychiatry 72:157-163.

Nithianantharajah J, Hannan AJ (2006) Enriched environments, experience-dependent plasticity and disorders of the nervous system. Nature reviews Neuroscience 7:697-709.

Nooner KB, Mennes M, Brown S, Castellanos FX, Leventhal B, Milham MP, Colcombe SJ (2013) Relationship of trauma symptoms to amygdala-based functional brain changes in adolescents. Journal of traumatic stress 26:784-787.

Nutt D, Demyttenaere K, Janka Z, Aarre T, Bourin M, Canonico PL, Carrasco JL, Stahl S (2007) The other face of depression, reduced positive affect: the role of catecholamines in causation and cure. Journal of psychopharmacology (Oxford, England) 21:461-471.

O'Doherty JP (2004) Reward representations and reward-related learning in the human brain: insights from neuroimaging. Current opinion in neurobiology 14:769-776.

Ochsner KN, Gross JJ (2005) The cognitive control of emotion. Trends in cognitive sciences 9:242-249.

Olff M, Van Zuiden M, Koch SBJ, Nawijn L, Frijling JL, Veltman DJ (2015) Intranasal oxytocin: miracle cure after trauma? European Journal of Psychotraumatology 6.

Organization WH (2015) Depression: Fact Sheet No 369. vol. 2015.

Paret C, Kluetsch R, Ruf M, Demirakca T, Hoesterey S, Ende G, Schmahl C (2014) Down-regulation of amygdala activation with real-time fMRI neurofeedback in a healthy female sample. Frontiers in behavioral neuroscience 8:299.

Pascucci T, Ventura R, Latagliata EC, Cabib S, Puglisi-Allegra S (2007) The medial prefrontal cortex determines the accumbens dopamine response to stress through the opposing influences of norepinephrine and dopamine. Cerebral cortex 17:2796-2804.

Patel R, Spreng RN, Shin LM, Girard TA (2012) Neurocircuitry models of posttraumatic stress disorder and beyond: a meta-analysis of functional neuroimaging studies. Neuroscience and biobehavioral reviews 36:2130-2142.

Patenaude B, Smith SM, Kennedy DN, Jenkinson M (2011) A Bayesian model of shape and appearance for subcortical brain segmentation. Neuroimage 56:907-922.

Pechtel P, Pizzagalli DA (2011) Effects of early life stress on cognitive and affective function: an integrated review of human literature. Psychopharmacology 214:55-70.

Perou R, Bitsko RH, Blumberg SJ, Pastor P, Ghandour RM, Gfroerer JC, Hedden SL, Crosby AE, Visser SN, Schieve LA, Parks SE, Hall JE, Brody D, Simile CM, Thompson WW, Baio J, Avenevoli S, Kogan MD, Huang LN (2013) Mental health surveillance among children--United States, 2005-2011. Morbidity and mortality weekly report Surveillance summaries (Washington, DC : 2002) 62 Suppl 2:1-35.

Pessoa L (2009) How do emotion and motivation direct executive control? Trends in cognitive sciences 13:160-166.

Pfeifer JH, Masten CL, Moore WE, 3rd, Oswald TM, Mazziotta JC, Iacoboni M, Dapretto M (2011) Entering adolescence: resistance to peer influence, risky behavior, and neural changes in emotion reactivity. Neuron 69:1029-1036.

Phelps EA, LeDoux JE (2005) Contributions of the amygdala to emotion processing: from animal models to human behavior. Neuron 48:175-187.

Philip NS, Sweet LH, Tyrka AR, Price LH, Bloom RF, Carpenter LL (2013) Decreased default network connectivity is associated with early life stress in medication-free healthy adults. European neuropsychopharmacology : the journal of the European College of Neuropsychopharmacology 23:24-32.

Piazza PV, Marinelli M, Rouge-Pont F, Deroche V, Maccari S, Simon H, Le Moal M (1996) Stress, glucocorticoids, and mesencephalic dopaminergic neurons: a pathophysiological chain determining vulnerability to psychostimulant abuse. NIDA research monograph 163:277-299.

Pine DS, Cohen E, Cohen P, Brook J (1999) Adolescent depressive symptoms as predictors of adult depression: moodiness or mood disorder? The American journal of psychiatry 156:133-135.

Pollak SD (2008) Mechanisms Linking Early Experience and the Emergence of Emotions: Illustrations From the Study of Maltreated Children. Current directions in psychological science 17:370-375.

Power JD, Barnes KA, Snyder AZ, Schlaggar BL, Petersen SE (2012) Spurious but systematic correlations in functional connectivity MRI networks arise from subject motion. NeuroImage 59:2142-2154.

Power JD, Schlaggar BL, Petersen SE (2015) Recent progress and outstanding issues in motion correction in resting state fMRI. NeuroImage 105:536-551.

Pryce CR, Dettling AC, Spengler M, Schnell CR, Feldon J (2004) Deprivation of parenting disrupts development of homeostatic and reward systems in marmoset monkey offspring. Biological psychiatry 56:72-79.

Qin S, Young CB, Supekar K, Uddin LQ, Menon V (2012) Immature integration and segregation of emotion-related brain circuitry in young children. Proceedings of the National Academy of Sciences of the United States of America 109:7941-7946.

Quirk GJ, Beer JS (2006) Prefrontal involvement in the regulation of emotion: convergence of rat and human studies. Current opinion in neurobiology 16:723-727.

Quirk GJ, Mueller D (2008) Neural mechanisms of extinction learning and retrieval. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 33:56-72.

Rabinak CA, Angstadt M, Welsh RC, Kenndy AE, Lyubkin M, Martis B, Phan KL (2011) Altered amygdala resting-state functional connectivity in post-traumatic stress disorder. Frontiers in psychiatry 2:62.

Reddy MS (2010) Depression: The Disorder and the Burden. Indian Journal of Psychological Medicine 32:1-2.

Rice K, Viscomi B, Riggins T, Redcay E (2014) Amygdala volume linked to individual differences in mental state inference in early childhood and adulthood. Developmental Cognitive Neuroscience 8:153-163.

Rincon-Cortes M, Sullivan RM (2014) Early life trauma and attachment: immediate and enduring effects on neurobehavioral and stress axis development. Frontiers in endocrinology 5:33.

Roy AK, Fudge JL, Kelly C, Perry JS, Daniele T, Carlisi C, Benson B, Castellanos FX, Milham MP, Pine DS, Ernst M (2013) Intrinsic functional connectivity of amygdala-based networks in adolescent generalized anxiety disorder. Journal of the American Academy of Child and Adolescent Psychiatry 52:290-299 e292.

Roy AK, Shehzad Z, Margulies DS, Kelly AM, Uddin LQ, Gotimer K, Biswal BB, Castellanos FX, Milham MP (2009) Functional connectivity of the human amygdala using resting state fMRI. NeuroImage 45:614-626.

Sah P, Faber ES, Lopez De Armentia M, Power J (2003a) The amygdaloid complex: anatomy and physiology. Physiological reviews 83:803-834.

Sah P, Faber ESL, Lopez De Armentia M, Power J (2003b) The Amygdaloid Complex: Anatomy and Physiology. Physiological Reviews 83:803-834.

Sanislow CA, Pine DS, Quinn KJ, Kozak MJ, Garvey MA, Heinssen RK, Wang PS, Cuthbert BN (2010) Developing constructs for psychopathology research: research domain criteria. J Abnorm Psychol 119:631-639.

Saylor CF, Finch AJ, Jr., Spirito A, Bennett B (1984) The children's depression inventory: a systematic evaluation of psychometric properties. Journal of consulting and clinical psychology 52:955-967.

Schiller D, Delgado MR (2010) Overlapping neural systems mediating extinction, reversal and regulation of fear. Trends Cogn Sci 14:268-276.

Seeley WW, Menon V, Schatzberg AF, Keller J, Glover GH, Kenna H, Reiss AL, Greicius MD (2007) Dissociable intrinsic connectivity networks for salience processing and executive control. The Journal of neuroscience : the official journal of the Society for Neuroscience 27:2349-2356.

Seo JH, Kuzhikandathil EV (2015) Dopamine D3 Receptor Mediates Preadolescent Stress-Induced Adult Psychiatric Disorders. PloS one 10:e0143908.

Shattuck DW, Leahy RM (2002) BrainSuite: an automated cortical surface identification tool. Medical image analysis 6:129-142.

Shaw P, Greenstein D, Lerch J, Clasen L, Lenroot R, Gogtay N, Evans A, Rapoport J, Giedd J (2006) Intellectual ability and cortical development in children and adolescents. Nature 440:676-679.

Shehzad Z, Kelly AM, Reiss PT, Gee DG, Gotimer K, Uddin LQ, Lee SH, Margulies DS, Roy AK, Biswal BB, Petkova E, Castellanos FX, Milham MP (2009) The resting brain: unconstrained yet reliable. Cerebral cortex 19:2209-2229.

Sheridan MA, Fox NA, Zeanah CH, McLaughlin KA, Nelson CA, 3rd (2012) Variation in neural development as a result of exposure to institutionalization early in childhood. Proceedings of the National Academy of Sciences of the United States of America 109:12927-12932.

Silk JS, Davis S, McMakin DL, Dahl RE, Forbes EE (2012) Why do anxious children become depressed teenagers? The role of social evaluative threat and reward processing. Psychological medicine 42:2095-2107.

Simmons AN, Arce E, Lovero KL, Stein MB, Paulus MP (2009) Subchronic SSRI administration reduces insula response during affective anticipation in healthy volunteers. The international journal of neuropsychopharmacology / official scientific journal of the Collegium Internationale Neuropsychopharmacologicum (CINP) 12:1009-1020.

Smoller JW (2016) The Genetics of Stress-Related Disorders: PTSD, Depression, and Anxiety Disorders. Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology 41:297-319.

Sobel RM, Markov D (2005) The impact of anxiety and mood disorders on physical disease: the worried not-so-well. Current psychiatry reports 7:206-212.

Spiegel D, Giese-Davis J (2003) Depression and cancer: mechanisms and disease progression. Biological psychiatry 54:269-282.

Sporns O (2011) The human connectome: a complex network. Annals of the New York Academy of Sciences 1224:109-125.

Sridharan D, Levitin DJ, Menon V (2008) A critical role for the right fronto-insular cortex in switching between central-executive and default-mode networks. Proceedings of the National Academy of Sciences of the United States of America 105:12569-12574.

Sripada RK, King AP, Welsh RC, Garfinkel SN, Wang X, Sripada CS, Liberzon I (2012) Neural dysregulation in posttraumatic stress disorder: evidence for disrupted equilibrium between salience and default mode brain networks. Psychosomatic medicine 74:904-911.

Stein DJ, Chiu WT, Hwang I, Kessler RC, Sampson N, Alonso J, Borges G, Bromet E, Bruffaerts R, de Girolamo G, Florescu S, Gureje O, He Y, Kovess-Masfety V, Levinson D, Matschinger H, Mneimneh Z, Nakamura Y, Ormel J, Posada-Villa J, Sagar R, Scott KM, Tomov T, Viana MC, Williams DR, Nock MK (2010) Cross-national analysis of the associations between traumatic events and suicidal behavior: findings from the WHO World Mental Health Surveys. PloS one 5:e10574.

Stevens JS, Jovanovic T, Fani N, Ely TD, Glover EM, Bradley B, Ressler KJ (2013) Disrupted amygdala-prefrontal functional connectivity in civilian women with posttraumatic stress disorder. Journal of psychiatric research 47:1469-1478.

Stevens WD, Spreng RN (2014) Resting-state functional connectivity MRI reveals active processes central to cognition. Wiley interdisciplinary reviews Cognitive science 5:233-245.

Sullivan R, Lasley EN (2010) Fear in love: attachment, abuse, and the developing brain. Cerebrum : the Dana forum on brain science 2010:17.

Tang Y, Kong L, Wu F, Womer F, Jiang W, Cao Y, Ren L, Wang J, Fan G, Blumberg HP, Xu K, Wang F (2013) Decreased functional connectivity between the amygdala and the left ventral prefrontal cortex in treatment-naive patients with major depressive disorder: a resting-state functional magnetic resonance imaging study. Psychol Med 43:1921-1927.

Taylor AE, Lee HE, Buisman-Pijlman FT (2014) Oxytocin treatment in pediatric populations. Frontiers in behavioral neuroscience 8:360.

Taylor KS, Seminowicz DA, Davis KD (2009) Two systems of resting state connectivity between the insula and cingulate cortex. Human brain mapping 30:2731-2745.

Taylor SF, Phan KL, Decker LR, Liberzon I (2003) Subjective rating of emotionally salient stimuli modulates neural activity. NeuroImage 18:650-659.

Thomas KM, Drevets WC, Whalen PJ, Eccard CH, Dahl RE, Ryan ND, Casey BJ (2001) Amygdala response to facial expressions in children and adults. Biological psychiatry 49:309-316.

Thomason ME, Dennis EL, Joshi AA, Joshi SH, Dinov ID, Chang C, Henry ML, Johnson RF, Thompson PM, Toga AW, Glover GH, Van Horn JD, Gotlib IH (2011) Resting-state fMRI can reliably map neural networks in children. NeuroImage 55:165-175.

Thomason ME, Marusak HA (2016) Toward understanding the impact of trauma on the early developing human brain. Neuroscience.

Thomason ME, Marusak HA, Tocco MA, Vila AM, McGarragle O, Rosenberg D (In Press) Altered Amygdala Connectivity in Youth Exposed to Trauma.

Thomason ME, Tocco MA, Quednau KA, Bedway AR, Carre JM (2013) Idle behaviors of the hippocampus reflect endogenous cortisol levels in youth. Journal of the American Academy of Child and Adolescent Psychiatry 52:642-652 e641.

Thompson RJ, Parker KJ, Hallmayer JF, Waugh CE, Gotlib IH (2011) Oxytocin receptor gene polymorphism (rs2254298) interacts with familial risk for psychopathology to predict symptoms of depression and anxiety in adolescent girls. Psychoneuroendocrinology 36:144-147.

Tottenham N (2012) Human amygdala development in the absence of species-expected caregiving. Developmental psychobiology 54:598-611.

Tottenham N (2013) The Importance of Early Experiences for Neuro-Affective Development. Current topics in behavioral neurosciences.

Tottenham N (2014) The importance of early experiences for neuro-affective development. Current topics in behavioral neurosciences 16:109-129.

Tottenham N, Hare TA, Millner A, Gilhooly T, Zevin JD, Casey BJ (2011) Elevated amygdala response to faces following early deprivation. Developmental science 14:190-204.

Tottenham N, Hare TA, Quinn BT, McCarry TW, Nurse M, Gilhooly T, Millner A, Galvan A, Davidson MC, Eigsti IM, Thomas KM, Freed PJ, Booma ES, Gunnar MR, Altemus M, Aronson J, Casey BJ (2010) Prolonged institutional rearing is associated with atypically large amygdala volume and difficulties in emotion regulation. Developmental science 13:46-61.

Tottenham N, Sheridan MA (2009) A review of adversity, the amygdala and the hippocampus: a consideration of developmental timing. Frontiers in human neuroscience 3:68.

Treadway MT, Zald DH (2011) Reconsidering anhedonia in depression: lessons from translational neuroscience. Neuroscience and biobehavioral reviews 35:537-555.

Tunnard C, Rane LJ, Wooderson SC, Markopoulou K, Poon L, Fekadu A, Juruena M, Cleare AJ (2014) The impact of childhood adversity on suicidality and clinical course in treatment-resistant depression. Journal of affective disorders 152-154:122-130.

Turner RJ, Noh S (1988) Physical disability and depression: a longitudinal analysis. Journal of health and social behavior 29:23-37.

Uddin LQ, Supekar KS, Ryali S, Menon V (2011) Dynamic reconfiguration of structural and functional connectivity across core neurocognitive brain networks with development. The Journal of neuroscience : the official journal of the Society for Neuroscience 31:18578-18589.

van den Heuvel MP, Mandl RC, Kahn RS, Hulshoff Pol HE (2009) Functionally linked resting-state networks reflect the underlying structural connectivity architecture of the human brain. Human brain mapping 30:3127-3141.

Van Dijk KR, Hedden T, Venkataraman A, Evans KC, Lazar SW, Buckner RL (2010) Intrinsic functional connectivity as a tool for human connectomics: theory, properties, and optimization. Journal of neurophysiology 103:297-321.

Van Dijk KR, Sabuncu MR, Buckner RL (2012) The influence of head motion on intrinsic functional connectivity MRI. NeuroImage 59:431-438.

van Wingen GA, Geuze E, Vermetten E, Fernandez G (2011) Perceived threat predicts the neural sequelae of combat stress. Molecular psychiatry 16:664-671.

Vuilleumier P, Armony JL, Driver J, Dolan RJ (2001) Effects of attention and emotion on face processing in the human brain: an event-related fMRI study. Neuron 30:829-841.

Waszczuk MA, Zavos HM, Gregory AM, Eley TC (2014) The phenotypic and genetic structure of depression and anxiety disorder symptoms in childhood, adolescence, and young adulthood. JAMA psychiatry 71:905-916.

Watson D, Clark LA (1984) Negative affectivity: the disposition to experience aversive emotional states. Psychological bulletin 96:465-490.

Watson D, Clark LA, Carey G (1988) Positive and negative affectivity and their relation to anxiety and depressive disorders. Journal of abnormal psychology 97:346-353.

Watson D, Clark LA, Weber K, Assenheimer JS, Strauss ME, McCormick RA (1995) Testing a tripartite model: II. Exploring the symptom structure of anxiety and depression in student, adult, and patient samples. J Abnorm Psychol 104:15-25.

Watson D, Tellegen A (1985) Toward a consensual structure of mood. Psychological bulletin 98:219-235.

Weisman O, Zagoory-Sharon O, Schneiderman I, Gordon I, Feldman R (2013) Plasma oxytocin distributions in a large cohort of women and men and their gender-specific associations with anxiety. Psychoneuroendocrinology 38:694-701.

Westlye LT, Walhovd KB, Dale AM, Bjornerud A, Due-Tonnessen P, Engvig A, Grydeland H, Tamnes CK, Ostby Y, Fjell AM (2010) Life-span changes of the human brain white matter: diffusion tensor imaging (DTI) and volumetry. Cerebral cortex 20:2055-2068.

Whalen P (1998) Fear, Vigilance, and Ambiguity: Initial Neuroimaging Studies of the Human Amygdala. Current directions in psychological science 7:177-188.

White MG, Bogdan R, Fisher PM, Munoz KE, Williamson DE, Hariri AR (2012) FKBP5 and emotional neglect interact to predict individual differences in amygdala reactivity. Genes, brain, and behavior 11:869-878.

Willner P (2005) Chronic mild stress (CMS) revisited: consistency and behavioural-neurobiological concordance in the effects of CMS. Neuropsychobiology 52:90-110.

Wismer Fries AB, Ziegler TE, Kurian JR, Jacoris S, Pollak SD (2005) Early experience in humans is associated with changes in neuropeptides critical for regulating social behavior. Proceedings of the National Academy of Sciences of the United States of America 102:17237-17240.

Wright P, Liu Y (2006) Neutral faces activate the amygdala during identity matching. Neuroimage 29:628-636.

Zald DH (2003) The human amygdala and the emotional evaluation of sensory stimuli. Brain research Brain research reviews 41:88-123.

Zannas AS, Binder EB (2014) Gene-environment interactions at the FKBP5 locus: sensitive periods, mechanisms and pleiotropism. Genes, brain, and behavior 13:25-37.

ABSTRACT

CHILDHOOD TRAUMA AND EMOTION PROCESSING NEUROCIRCUITRY

by

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Advisor: Dr. Moriah E. Thomason

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Childhood trauma is one of the strongest risk factors for a range of common and debilitating neuropsychiatric disorders, including anxiety, depression, and posttraumatic stress disorder (PTSD). These emotion-related disorders have their roots in childhood and adolescence, underscoring a critical need to understand their biological bases in early life. In this dissertation, we evaluate how childhood trauma impacts emotion processing neurocircuitry in a sample of high-risk urban youth, ages 7-15. In four inter-related studies, we test neural function and functional connectivity of core emotion processing regions, including the amygdala, insula, and pregenual/subgenual anterior cingulate cortex (pgACC/sgACC). To examine the relevance of observed neurological changes, we evaluate behavioral performance on emotion processing neuropsychological tasks, as well as specific dimensions of subjective affective experience.

Results indicate that, relative to matched comparison youth, trauma-exposed youth have (1) increased neural response to salient emotional cues in amygdala and insula, (2) reduced functional connectivity between amygdala and pgACC/sgACC, a pathway critical for emotion regulation, and (3) altered within- and between-network connectivity of the salience network, involved in detecting and orienting attention to salient emotional stimuli. These neurological

changes are accompanied by behavioral alterations: trauma-exposed youth have a lower ability to ignore distracting emotional information, and to automatically regulate emotion. Additionally, observed neurobehavioral changes relate to a specific dimension of affective experience – reward sensitivity (RS), rather than negative affect. Moreover, trauma-exposed youth with the greatest neurobehavioral impairment report lower RS, suggesting reduced positive environmental engagement.

These results suggest that RS may be a marker of stress susceptibility, a notion supported by emerging basic and clinical research. Based on our neurobehavioral findings, we discuss potential implications for intervention, and relay an emerging framework that dissociates neurological effects of different trauma types (i.e., threat/victimization vs. deprivation/neglect). In closing, we discuss future directions, including longitudinal research and evaluating the modulation of learned fear – a neurobehavioral mechanism that depends on emotion processing neurocircuitry, but has yet to be tested in trauma-exposed youth.

AUTOBIOGRAPHICAL STATEMENT

For as long ago as I can remember, I have been interested in the intersection of behavior and biology. This interest narrowed to the field of neuroscience when I recognized the dramatic and sometimes devastating consequences that brain-based disorders can have on human behavior, health, and wellbeing. By shadowing psychiatrists and neuroscientists at the UCLA Mood Disorders Research Program, I was fortunate to witness firsthand the clinical, behavioral, and neurological manifestation of affective disorders. I was fascinated by the fact that these common and debilitating disorders are biologically rooted in brain circuitry.

The importance of understanding the biological bases of affective disorders became all the more lucid and urgent to me during my senior year of college. A classmate and friend, whose depression was unknown to most all, took his own life just months before our college graduation. I am grateful to be receiving the training I need to become an affective neuroscientist striving to better understand and prevent these insidious disorders from taking root in young people.

In memory of Noah.