Early Life Stress, Mood, and Anxiety Disorders

Shariful A. Syed[1] and Charles B. Nemeroff[1]

Abstract

Early life stress has been shown to exert profound short- and long-term effects on human physiology both in the central nervous system and peripherally. Early life stress has demonstrated clear association with many psychiatric disorders including major depression, posttraumatic stress disorder, and bipolar disorder. The Diagnostic and Statistics Manuel of Mental Disorders (DSM) diagnostic categorical system has served as a necessary framework for clinical service, delivery, and research, however has not been completely matching the neurobiological research perspective. Early life stress presents a complex dynamic featuring a wide spectrum of physiologic alterations: from epigenetic alterations, inflammatory changes, to dysregulation of the hypothalamic pituitary axis and has further added to the challenge of identifying biomarkers associated with psychiatric disorders. The National Institute of Mental Health's proposed Research Domain Criteria initiative incorporates a dimensional approach to assess discrete domains and constructs of behavioral function that are subserved by identifiable neural circuits. The current neurobiology of early life stress is reviewed in accordance with dimensional organization of Research Domain Criteria matrix and how the findings as a whole fit within the Research Domain Criteria frameworks.

Background

Early life stress (ELS) comprised of various forms of child abuse and neglect has been shown to exert profound short- and long-term effects on human physiology both in the central nervous system (CNS) and peripherally.[1] ELS has clearly been demonstrated to be associated with increased risk for many psychiatric disorders including major depression, posttraumatic stress disorder (PTSD), and bipolar disorder.[2] The past decade has witnessed an explosion of findings in relation to the consequences of adverse early life experience.

Grossly, these findings pertain to a wide spectrum of physiologic alteration: from epigenetic alterations, inflammatory changes to dysregulation of the hypothalamic pituitary axis (HPA) axis. ELS presents a complex dynamic which from a neurobiological research perspective does not completely match the DSM-defined categorical diagnoses.

DSM diagnostic categories, a necessary framework for clinical service delivery and research, clearly combine pathophysiologically distinct profiles in a single diagnostic category, which may be a contributing factor to inconsistency in research findings across investigations,[3] including the effort to identify diagnostic biomarkers associated with mental illnesses.[4]

National Institute of Mental Health's proposed Research Domain Criteria (RDoC) initiative incorporates a dimensional approach to assess discrete domains and constructs of behavioral functions that are subserved by an identified neural circuit. The five initial domains being: negative valence (acute threat, anxiety, and sustained threat), positive valence, cognitive systems, social processes, and arousal/modulatory systems. Each construct is examined along seven units of analyses: genes,

[1]Department of Psychiatry and Behavioral Sciences, Miller School of Medicine, University of Miami, Miami, FL, USA

Corresponding author:
Charles B. Nemeroff, Department of Psychiatry and Behavioral Sciences, Miller School of Medicine, University of Miami, 1120 Northwest 14th Street, Suite 1455, Miami, FL 33136, USA.
Email: cnemeroff@med.miami.edu

molecules, cells, circuits, physiology, behavior, self-reports, and paradigms.[5]

Within each domain are constructs that significantly correlate with hallmark features of psychiatric illnesses such as negative valence of PTSD. This is a novel approach which has clear virtues but several potential drawbacks as we have noted previously.[6]

Given the broad scope of neurobiological findings from investigations of ELS, it may be argued that this dimensional/matrix approach will be informative for elucidating how ELS mediates increased risk for psychiatric illness. We review the current neurobiology of ELS in accordance with the dimensional organization of the RDoC matrix in mind. We then comment on how these findings as a whole fit within the RDoC framework, the tenets it was founded upon, and objectives it was designed to achieve.

Genes to Circuits: Corticotropin-Releasing Factor's Role in Stress Response Neurobiology

There is a well-established link between early adverse life experience and the development of psychiatric disorders, of which the neurobiological stress response is believed to play a seminal role. The two main components of the mammalian stress response are the sympathetic adreno-medullary (SAM) system and HPA axis.[7] Both are modulated by CNS circuits involving areas of the prefrontal cortex, hippocampus, amygdala, hypothalamic, and brain stem nuclei.

Corticotropin-releasing hormone (also referred to as CRH) producing neurons oversee the entire mammalian stress response, coordinating the autonomic, endocrine, immune, and behavioral responses to stress.[8] The highest concentrations of Corticotropin-releasing factor (CRF) are found in the parventricular nucleus (PVN) of the hypothalamus, which primarily regulates the neuroendocrine stress response.[9] CRF-producing neurons located in the central nucleus of the amygdala are involved in processing emotional stress responses and the SAM response as well.

As regards this SAM response the CRF neurons in the central nucleus of the amygdala project to the CRF neurons in locus coeruleus norepinephrine (NE) cells which project to the lateral thalamus leading to subsequent activation of the sympathetic preganglionic neurons that stimulate release of epinephrine from the adrenal medulla. Central nucleus of amygdala CRF cells are involved in stress-induced activation of the HPA axis,[10] using an indirect pathway through the bed nucleus of the stria terminalis (BNST), where CRF neuron projections innervate the PVN neurons of hypothalamic.[11–13] Following activation of HPA axis, CRF is released from the PVN to the hypothalamo-hypophysial portal circulation from nerve terminals in the median eminence where it stimulates adrenocorticotropin hormone (ACTH) release from the anterior pituitary.

ACTH in turn stimulates release of glucocorticoids (GCs) from the adrenal cortex.[14] Able to permeate the blood—brain barrier, GCs reduce activation of the HPA axis via stimulation of GC receptors (GRs) within the hippocampus, hypothalamus, and anterior pituitary.[15] The critical role of amygdalar CRF has brought to attention the wide spread CRF receptors located in brain and their converging pathways in orchestrating stress reactions.[16,17]

Two G protein-linked subtypes of CRF receptors CRF1 and CRF2 have been found in the anterior pituitary as well as in subcortical and cortical brain areas.[18,19] In general, the stress response appears to be mediated by CRF1 receptors, whereas CRF2 receptor activation appears to diminish the stress response.

The response to psychosocial stress, of which ELS likely represents a specific subtype, also involves "higher appraisal" by cortical and subcortical regions of brain containing CRF1 receptors, namely, cingulate cortex, orbital/medial prefrontal cortex, and hippocampus;[20] all these areas comprise part of the converging pathways described earlier.

Much evidence points to the role for CRF as a neurotransmitter coordinating immune, autonomic, endocrine, and behavioral stress responses, supported by the finding that CRF1 receptors are more abundant in cortico-limbic pathways that mediate fear- and anxiety-related behaviors.[21]

In laboratory animals, CRF administration directly into the CNS leads to activation of the autonomic nervous system, elevation of peripheral catecholamines, and increased heart rate and blood pressure. CNS administration of CRF has been shown to induce diminished food intake, disturbed sleep patterns, facilitation of fear conditioning, and increased startle response—behaviors that parallel symptoms of depressive/anxiety disorders. In non-human primates, direct administration of CRF into the CNS produced depressive symptoms including huddling behavior and inactivity.[22]

Interestingly, CRF antagonists have been shown to attenuate the anxiety and depressive symptoms mediated by CNS administration of CRF as well as to possess intrinsic anxiolytic properties in a variety of preclinical paradigms.[23,24]

In humans, cerebrospinal fluid (CSF) CRF concentrations are elevated in drug-free depressed patients compared with controls.[25–27] The literature supports CRF as a key mediator of the stress response and dysregulation of the CRF system; which may in part explain the heightened vigilance and enhanced startle observed in patients with anxiety and mixed depression–anxiety. ELS mediates persistent neurodevelopmental changes through alterations to the CRF and the HPA axis, which in this

review will be shown to relate to the pathophysiology of mood and anxiety disorders.

Early Life Stress and the HPA Axis: Genes, Molecules, Cells, Physiology, and Circuits

Genes

It has become increasingly established that genetics contribute to the risk for development of major psychiatric disorders. In addition, ELS in the form of child abuse and neglect serve as important risk factor for psychiatric disorders.[28,29] One of the goals of RDoC is to discover gene candidates that can function as markers to predict individual disease risk, prognosis, and treatment responses. Single-nucleotide polymorphisms (SNPs) have been investigated to evaluate for their interactions with early life experience and effect on higher dimensions of neurobiological processes.

A novel approach that has been utilized in recent studies tests the hypothesis that gene variants may modulate the effect of ELS on the longitudinal risk for mental illness. Diathesis–stress theories of depression suggest that individual's sensitivity to stressful events depends, in part, on their genotype.[30,31] Investigations to date have largely supported this theory, with many studies demonstrating gene × environment (G × E) interactions that predict both mental disorder risk and treatment outcomes in humans. To date, there have been a handful of genetic polymorphisms proposed that shall be reviewed here: 5HTTLPR, Monoamine Oxidase A (MAOA), FKBP5, CRHR1, Brain-Derived Neurotrophic Factor (BDNF), and OPRL1.

Serotonin transporter polymorphism. Evidence continues to increasingly suggest that the pathophysiology and treatment response for a subset of individuals with depression may be influenced by a polymorphism in the promoter region of the serotonin transporter gene (SERT).

The SERT is encoded in humans by a single gene (SLC6A4) located on chromosome 17q11.1-q12. A functional three base pair repeat polymorphism has been identified in the promoter region of this gene and is also known as 5-HTTLPR. The short variant of the polymorphism is denoted as "*s*" and long variant as "*l*." The "*s/s*" or "*s/l*" genotype is associated with reduced transcription of the SERT gene and reduced 5-HT uptake in comparison to the "*l/l*" genotype.[32–35]

The *s* allele of the SERT genotype has been associated in some studies with a predisposition toward development of depression in humans. However, not all individuals with *s/s* or *s/l* develop depression. Environmental factors have been increasingly shown to interact with SERT genotypes to increase individual likelihood of developing depression in adulthood. Mehta et al.[36] found, in a non-psychiatric cohort, that *s* allele carrier

status predicted late post partum depressive symptoms only in the presence of negative life events.

Multiple studies have shown G × E interactions linking ELS to an increased risk of depression. In non-human primates, Coplan et al.[37] have demonstrated that ELS in the form of variable foraging demand leads to elevated CSF CRF concentrations, most notably in subjects with the *s/s* and *s/l* 5HTTLPR genotype. Human studies have also revealed HPA axis hyperactivity in patients who reported a history of ELS and current depression.[38]

Barr et al.[39] assessed the influence of rearing condition and the rh5-HTTLPR polymorphism on ACTH release. The *s* allele coupled with ELS led to increase plasma ACTH concentrations, confirming the findings of Coplan et al. cited previously. Much research has focused on the interaction between SERT polymorphisms, ELS, and depression. Caspi et al.[40] were the first to demonstrate an association between depression, ELS, and the 5-HTTLPR genotype. Individuals exposed to childhood maltreatment, possessing the *s/s* genotype were shown to have the highest probability of developing a major depressive episode, followed by the *s/l* genotype. In a general population study, a three-way interaction among childhood abuse × adult traumatic experience × *s* allele carrier status was found to be associated with higher Beck Depression Inventory-II (BDI-II) scores.[41] A meta-analysis by Karg et al.[42] found strong evidence supporting the association between childhood maltreatment and the *s* allele and increased stress sensitivity.

Interestingly, subjects with the *l/l* genotype did not show an increased risk of developing major depressive episodes even in the presence of ELS. In the wake of this paradigm shifting study, numerous studies have addressed and confirmed these findings.[43]

This G × E discovery leads to the interesting question, namely, whether we can use a patient's genotype for the SERT as well as other polymorphisms coupled with a history of ELS as criteria for early intervention to prevent the development of major depression in vulnerable individuals.

Studies of the interaction of SERT polymorphisms and ELS in children also point to the value of early intervention as a strategy for children with a known history of abuse. Kaufman et al.[44] showed that a supportive environment seemed to protect children with the *s/s* genotype and a history of maltreatment from developing depression. Recent studies have demonstrated G × G × E interaction including the SERT gene.

MAOA. Childhood maltreatment may be the most common form of ELS in western society and is associated with a wide array of mental health outcomes.[45] Although the risk for developing conduct disorder, antisocial personality disorder, and criminal behavior is increased by ELS, most children do not develop into adult criminals.[46]
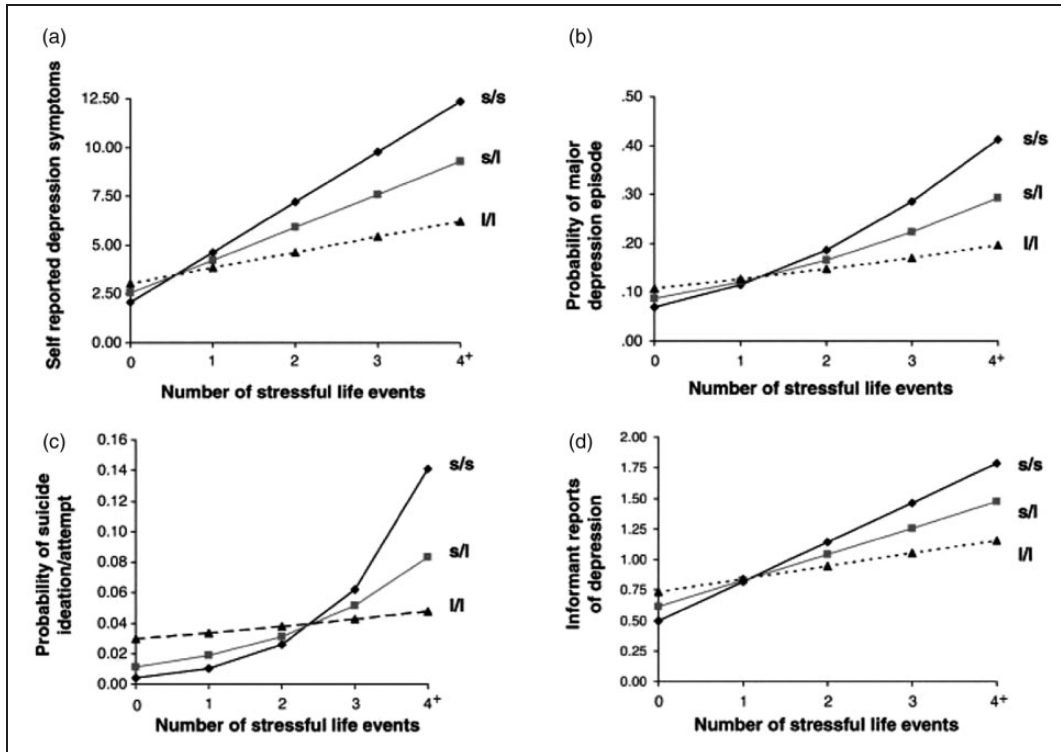
Figure 1. Influence of life stress on depression: Moderation by a polymorphism in the 5-HTT gene. Results of multiple regression analyses estimating the association between number of stressful life events (between ages 21 and 26 years) and depression outcomes at age 26 as a function of 5-HT T genotype. Among the 146 s/s homozygotes, 43 (29%), 37(25%), 28 (19%), 15 (10%), and 23 (16%) study members experienced zero, one, two, three, and four or more stressful events, respectively. Among the 435 s/l heterozygotes, 141 (32%), 101 (23%), 76 (17%), 49 (11%), and 68 (16%) experienced zero, one, two, three, and four or more stressful events. Among the 264 l/l homozygotes, 79 (29%), 73 (28%), 57 (21%), 26 (10%), and 29 (11%) experienced zero, one, two, three, and four or more stressful events. (a) Self-reports of depression symptoms. The main effect of 5-HT TLPR (i.e., an effect not conditional on other variables) was marginally significant ($b = -0.96$, $SE = 0.52$, $t = 1.86$, $p = 0.06$), the main effect of stressful life events was significant ($b = 1.75$, $SE = 0.23$, $t = 7.45$, $p < 0.001$), and the interaction between 5-HT TLPR and life events was in the predicted direction ($b = -0.89$, $SE = 0.37$, $t = 2.39$, $p = 0.02$). The interaction showed that the effect of life events on self-reports of depression symptoms was stronger among individuals carrying an s allele ($b = 2.52$, $SE = 0.66$, $t = 3.82$, $p < 0.001$ among s/s homozygotes, and $b = 1.71$, $SE = 0.34$, $t = 5.02$, $p < 0.001$ among s/l heterozygotes) than among l/l homozygotes ($b = 0.77$, $SE = 0.43$, $t = 1.79$, $p = 0.08$). (b) Probability of major depressive episode. The main effect of 5-HT TLPR was not significant ($b = -0.15$, $SE = 0.14$, $z = 1.07$, $p = 0.29$), the main effect of life events was significant ($b = 0.37$, $SE = 0.06$, $z = 5.99$, $p < 0.001$), and the G 3 E was in the predicted direction ($b = -0.19$, $SE = 0.10$, $z = 1.91$, $p = 0.056$). Life events predicted a diagnosis of major depression among s carriers ($b = 0.52$, $SE = 0.16$, $z = 3.28$, $p = 0.001$ among s/s homozygotes, and $b = 0.39$, $SE = 0.09$, $z = 4.24$, $p < 0.001$ among s/l heterozygotes) but not among l/l homozygotes ($b = 0.16$, $SE = 0.13$, $z = 1.18$, $p = 0.24$). (c) Probability of suicide ideation or attempt. The main effect of 5-HT TLPR was not significant ($b = -0.01$, $SE = 0.28$, $z = 0.01$, $p = 0.99$), the main effect of life events was significant ($b = 0.51$, $SE = 0.13$, $z = 3.96$, $p < 0.001$), and the G 3 E interaction was in the predicted direction ($b = -0.39$, $SE = 0.20$, $t = 1.95$, $p = 0.051$). Life events predicted suicide ideation or attempt among s carriers ($b = 0.48$, $SE = 0.29$, $z = 1.67$, $p = 0.09$ among s/s homozygotes, and $b = 0.91$, $SE = 0.25$, $z = 3.58$, $p < 0.001$ among s/l heterozygotes) but not among l/l homozygotes ($b = 0.13$, $SE = 0.26$, $z = 0.49$, $p = 0.62$). (d) Informant reports of depression. The main effect of 5-HT TLPR was not significant ($b = -0.06$, $SE = 0.06$, $t = 0.98$, $p = 0.33$), the main effect of life events was significant ($b = 0.23$, $SE = 0.03$, $t = 8.47$, $p < 0.001$), and the G 3 E was in the predicted direction ($b = -0.11$, $SE = 0.04$, $t = 2.54$, $p < 0.01$). The effect of life events on depression was stronger among s carriers ($b = 0.39$, $SE = 0.07$, $t = 5.23$, $p < 0.001$ among s/s homozygotes, and $b = 0.17$, $SE = 0.04$, $t = 4.51$, $p < 0.001$ among s/l heterozygotes) than among l/l homozygotes ($b = 0.14$, $SE = 0.05$, $t = 2.69$, $p < 0.01$). From Caspi et al.[40] Reprinted with permission from AAAS.

Caspi et al.[40] were among the first to suggest that individual differences at a functional polymorphism in the promoter region of the MAOA gene may modulate children's response to maltreatment (Figure 1).

Located on the X chromosome (Xp11.123–11.4),[48] it encodes the MAOA enzyme, which metabolizes neurotransmitters including NE, serotonin (5-HT), and dopamine.[49] Genetic deficiencies of MAOA have been linked to the development of behavior disturbances in children. SNPs of MAOA demonstrated significant G × E interaction; lower MAOA activity showed a stronger effect of childhood maltreatment to positively predict

the percentage of subjects that fit diagnostic criteria for conduct disorder. Conversely, males with high MAOA activity did not show significantly increased risk for conduct disorder.

Kim-Cohen et al., using a representative birth cohort sample of 7-year-old boys, confirmed and extended Caspi et al.'s original findings. They showed a G × E interaction in which boys with the low-activity MAOA allele had mental health problems scores that were half a standard deviation higher than boys with the high-activity allele.[50] Further, their meta-analysis results corroborated this G × E effect.

Given that MAOA metabolizes neurotransmitters that are central to multiple brain functional circuits associated with stress regulation, it represents one of the factors involved in biological sensitivity to life stress.[51,52]

FKBP5. G × E research has also spurred the investigation of PTSD, in an attempt to answer the quandary of how some individuals are more likely than others to develop a stress disorder when exposed to similar levels of trauma.[53–56]

It has becoming clear that there are predisposing genetic and environment factors contributing to an individual risk after experiencing trauma.[57] There is, of course, also the dimension of resilience that serves to protect individuals exposed to trauma from developing psychological sequelae. The association between child abuse and adult PTSD has been well established.[58] Given that PTSD is strongly associated with long-lasting alterations in HPA axis sensitivity and increased GR sensitivity, a natural extension of G × E research has examined whether HPA axis gene candidates mediate the increased susceptibility to PTSD after ELS.[27,59]

FKBP5 is a gene that encodes a chaperone protein that directly interacts with the GR-heterocomplex allowing it to translocate into the nucleus and interact with GRs. Overexpression of FKBP5 mediates a reduction in GC signal transduction, leading to reduced efficiency of GR signaling required for negative feedback accompanied by relative increases in plasma cortisol[60]; this HPA axis configuration matches very well what is seen in PTSD.[27]

Four FKBP5 SNPs were found to significantly predict the PTSD Symptom Score in individuals with a history of child abuse. All four SNPs have been associated with the presence of higher levels of FKBP5, consistent with the physiological mechanisms mediating GC sensitivity.[61]

Thus, in addition to FKBP5 polymorphisms interacting with child abuse to predict levels of adult PTSD symptoms, it is believed that FKBP5 alleles may enhance the effect of acutely released cortisol leading to abnormal FKBP5 expression driving persistent disturbances of GR sensitivity.[62] Zannas and Binder[63] found that exposure to child abuse leads to significant demethylation of CpG in the functional GRE of FKBP5 gene mediating

GR resistance. Klengel et al.[64] reported this demethylation as linked to an ELS-dependent increased FKBP5 gene transcription with subsequent long-term dysregulation of the stress hormone system (Figure 2). McGowan et al.[65] showed that suicide victims with reported ELS demonstrated increased cytosine methylation of the NR3C1 promoter of GR gene.

CRHR1/OPRL1/BDNF. Although the SERT has received much attention, it is clear that several genes moderate vulnerability to depression.[66] Moreover, given that increased activity of HPA axis, in part due to CRH neuronal hyperactivity,[7] has been demonstrated in Major Depressive Disorder (MDD), genes regulating HPA axis physiology in general, and those of the various components of the CRH system in particular, have been implicated in the regulation of stress reactivity.[67,68]

Studies have also demonstrated a significant association between ELS and CRF receptor activity (CRF-R1 or CRHR1). Clinical studies of depressed patients have revealed both increased CSF CRF concentrations and CRF-R1 mRNA expression in limbic brain regions including the amygdala.[69,70] Bangasser et al.[71] showed a sex difference in CRF receptor cellular signaling and receptor internalization, which rendered females more sensitive to CRF. Another study demonstrated a sex-specific association, showed that a specific SNP of the pituitary adenylate cyclase-activating polypeptide predicted PTSD diagnosis and symptoms in females only.[72]

Bradley et al. demonstrated that genetic variants of the CRHR1 moderate the effect of child abuse on adult depressive symptoms. Laucht et al.[73] found that the impact of childhood maltreatment on adult depressive symptoms was higher in individuals with two copies of the CRHR1 TAT haplotype. A haplotype of three SNPs in intron 1 of the CRHR1 gene was associated with a diminished effect of child abuse on adult depressive symptoms.[74] Thus, a genotype/haplotype may serve as a predictor both risk/resilience in those with history of child abuse and neglect.

In contrast to the *s* variant of 5HTTLPR, the discovered CRHR1 haplotype is not considered a functional variant. Rather it is believed that CRHR1 SNPs may modulate gene transcriptional activity and be in linkage disequilibrium with a functional variant.

This has led to the inclusion of gene × gene interactions in addition to more classical G × E studies to elucidate genetic contribution to depression risk.

Thus, Ressler et al. found that variants in the 5-HTTLPR interact with CRHR1 genotypes to predict current adult depressive symptom. Individuals carrying a "risk" allele in both genes demonstrated more severe depressive symptom at lower levels of child abuse.[75] Interestingly, this interaction was present only when the measure of child abuse was stratified across three severity
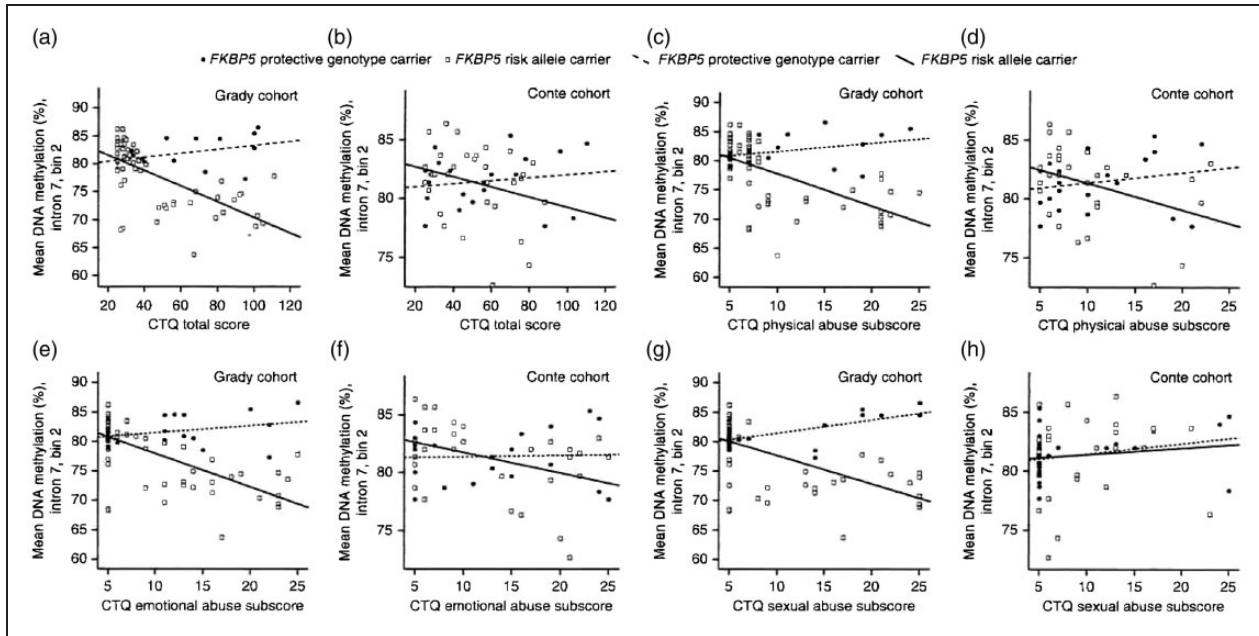
Figure 2. Differential FKBP5 intron 7 DNA methylation depends on genotype and trauma exposure. Correlation between intron 7 bin 2, mean methylation, and log-transformed CTQ scores by FKBP5 rs1360780 genotype in the Grady and Conte cohort are shown. (a) Grady cohort. Risk allele carriers exhibited a strong negative correlation ($R = 0.646$, $p < 0.001$) between methylation and CTQ total load compared with carriers of the protective genotype ($R = 0.414$, $p = 0.078$; Fisher Z score $= 4.23$, $p < 0.001$). (b) Conte cohort. Correlation between methylation and total CTQ in risk allele carriers ($R = 0.273$, $p = 0.124$) and in carriers of the protective genotype ($R = 0.153$, $p = 0.485$; Fisher Z score $= 1.5$, $p = 0.133$). (c) Grady cohort. Negative correlation was found between methylation and the CTQ physical abuse subscore in risk allele carriers ($R = 0.586$, $p < 0.001$) but not in carriers of the protective genotype ($R = 0.360$, $p = 0.130$; Fisher Z score $= 4.49$, $p < 0.001$). (d) Conte cohort. Negative correlation was observed between methylation and the CTQ physical abuse subscore in risk allele carriers ($R = 0.397$, $p = 0.022$) but not in carriers of the protective genotype ($R = 0.246$, $p = 0.258$; Fisher Z score $= 2.33$, $p = 0.019$). (e) Grady cohort. Negative correlation was found between methylation and the CTQ emotional abuse subscore in risk allele carriers ($R = 0.685$, $p < 0.001$) but not in carriers of the protective genotype ($R = 0.321$, $p = 0.181$; Fisher Z score $= 4.1$, p < 0.001). (f) Conte cohort. Negative correlation was found between methylation and the CTQ emotional abuse subscore in risk allele carriers ($R = 0.397$, $p = 0.022$) but not in carriers of the protective genotype ($R = 0.022$, $p = 0.922$; Fisher Z score $= 1.53$, $p = 0.126$). (g) Grady cohort. Negative correlation was found between methylation and the CTQ sexual abuse subscore in risk allele carriers ($R = 0.656$, $p < 0.001$) but not in carriers of the protective genotype ($R = 0.599$, $p = 0.007$; Fisher Z score $= 5.17$, $p < 0.001$). (H) Conte cohort. Negative correlation was found between methylation and the CTQ sexual abuse subscore in risk allele carriers ($R = 0.118$, $p = 0.514$) and in carriers of the protective genotype ($R = 0.305$, $p = 0.922$; Fisher Z score $= 0.68$, $p = 0.496$). From Klengel et al.[64] Reprinted by permission from Macmillan Publishers.
CTQ: Childhood Trauma Questionnaire.

levels. This highlights the importance of defining ELS in the investigation of G × E interactions. Furthermore, these findings support the plethora of research that has suggested a dose-response relationship between life stressors and risk of behavioral problems.[45,76,77]

This level of analysis is consistent with the fact that early adverse experience impacts both CRF and 5-HT systems suggesting significant interpermeation of the two neural circuits. These interconnections supported by studies that show that Pre-Frontal Cortex (PFC), hippocampus, and basolateral amygdala receive significant serotonergic innervation which is influenced by CRF-mediated increase Gamma-Aminobutyric acid (GABA) ergic inhibition of 5-HT at the dorsal raphe nuclei.[78,79]

Another G × G interaction with implications of vulnerability to depression is between SLC6A4 and BDNF. Meta-analyses have suggested that alteration in serotonergic activity may serve as a prodrome for later changes in neural plasticity of which BDNF is essential.[80,81]

One study suggested that the BDNF Met allele may serve as a protection against the adverse effects associated with the 5-HTTLPR *s* allele in healthy individuals. However, in maltreated children, the combination of BDNF Met with 5-HTTLPR *s* allele was associated with an increased risk for depression.[82] Given that BDNF is known to exert a direct effect on neuronal growth and plasticity in hippocampus and amygdale,[83–85] it comes as little surprise to find that BDNF

Val66Met × ELS interacts to predict significant changes to brain structure and function.[86] Early life maltreatment in rats showed persistent changes in methylation of BDNF, altering adult BDNF gene expression in prefrontal cortex.[87]

To add to the ELS-HPA axis gene interaction story, a SNP found in the opioid receptor like 1 (Oprl1) gene in patients with PTSD symptoms after a traumatic event is associated with a self reported history of childhood trauma.[88] The same SNP is association with altered fear learning and fear discrimination mechanisms including differential amygdala–insula functional connectivity which has been linked to PTSD.[89]

Molecules, Cells, and Physiology

Given the central role of the HPA axis and CRF in mediating the endocrine, immune, behavioral, and autonomic effects of stress, both preclinical and clinical studies have explored and confirmed that ELS does indeed produce persistent changes that correlate with increase risk of development of psychiatric illness.

Preclinical studies. Many of the initial studies used various forms of maternal separation in rats as a model of ELS, a species in which much of the neural development in brain occurs in the post-natal period. Alterations in maternal care between dams and their pups have already been shown to set into motion molecular events that directly pertain to regulation of the HPA axis, namely, that maternal licking and grooming regulate methylation of the GR gene in hippocampus.[90] GR genes, obviously, directly influence how many receptors are present on hippocampal neurons; increase receptor number allows for efficient termination of the stress response and protects against chronic effects of allostatic load.[91]

Brief maternal deprivation during the neonatal period in rats resulted in significant increases of basal and stress-induced ACTH concentrations, CRF concentrations in median eminence, as well as reduced CRF-R1 receptor density in the anterior pituitary.[92]

Further, prolonged maternal separation are associated with long-term changes in adult male rats, demonstrating increased CSF CRF concentrations as well as increased CRF mRNA expression in the PVN, central nucleus of the amygdala, BNST, and locus coeruleus.[93]

Similarly, ELS in rats is associated with the disruption of the negative feedback of the HPA axis, with maternally deprived rats escaping from suppression of plasma ACTH and corticosterone by dexamethasone.[94]

Studies of maternal grooming of rats showed that those that received less licking and grooming were found to have shorter dendritic branches and lower spine densities in CA1 cells which was associated with impaired hippocampal long-term potentiation as well as concurrent reductions of GC and MR receptor density.[95]

Studies in the non-human primate have repeatedly shown that ELS paradigms mediate persistent neuroendocrine, neurotransmitter, and behavioral effects.[37,96] In the bonnet macaque, alteration of food availability to the mother–infant dyad known as variable foraging demand has been repeatedly associated with marked and persistent increases in CSF CRF concentrations.[97]

The resulting neglectful maternal care of infant primates persists into their adulthood, as they have been show to be more fearful, exhibit greater weight gain, and decreased glucose disposal rates.[98,99] In Rhesus monkeys, repeated maternal separation was associated with a flattened diurnal secretion pattern of cortisol and increased acoustic startle reactivity and cortisol reactivity to separation.[100]

Clinical studies. The influence of ELS on HPA axis activity in humans has been an area of extensive and closely scrutinized research, in part due to the fact that studies in this area have shown child abuse and neglect is associated with both increased and decreased HPA axis activity.

Using various models validated human stress, such as the Trier Social Stress Test and the combined dexamethasone CRF stimulation test, HPA axis hyperactivity was demonstrated in depressed women and men with ELS as demonstrated by increases in both the ACTH and cortisol response as well as increased CSF CRF concentrations.[38,101–103] Increased basal and post stress cortisol levels were reported in patients with major depression and borderline personality disorder who reported a history of childhood trauma.[104]

In contrast, individuals with ELS in the form of child abuse have been reported to exhibit reduced basal cortisol levels as well as blunted cortisol response to provocative stimuli.[105] Likewise, ELS is well documented to increase the risk for development of PTSD, which is characterized by an "endocrine signature" of GR hypersensitivity and reduced cortisol signaling.[106]

These described discordant findings fuels active investigation into the precise effects of ELS. A recent study has attempted to reconcile them by suggesting a two pathway model in which ELS invokes interactions of the GC system with oxytocin and serotonergic systems to mediate an ultimate outcome of hyper- or hypoactivity of the HPA axis.[107]

Briefly, oxytocin is known to mediate attachment, social affiliation, intimacy, trust, and has recently been shown to be affected by ELS. Exposure to maltreatment in childhood was significantly inversely associated with CSF oxytocin concentrations.[108] Furthermore, SNP variation of the oxytocin receptor interacted with ELS to predict anxiety and depression severity.[109]

Early Life Stress and Brain Circuits

The RDoC was initially conceptualized as a circuit-based framework for the study of psychiatric illness, the timing of its emergence coincide with the rapidly developing understanding of brain mechanisms mediating complex behaviors, which has been increasingly blurring the line between the clinical disciplines of psychiatry and neurology.[110]

Because the pathogenesis of depression is most appropriately understood as a multidimensional, system-level disorder involving functionally overlapping pathways, the need for biological algorithms to define homeostatic emotional control during stress is of paramount importance.[111]

That ELS produces persistent increases in CSF CRF neural circuits, a hallmark of HPA axis hyperactivity, and dysregulation of cortico–limbic circuits puts it in a position of fundamental importance in exploring pathogenic mechanisms that may underlie major psychiatric illnesses such as major depression and PTSD.

Recent advances in structural and functional brain imaging have begun to elucidate the long-lasting effects of childhood maltreatment on the CNS.[112] Moreover, the use of imaging genetics has also revealed the importance of selective polymorphisms of candidate genes as modulating regional brain activity.

Within the context of ELS, emerging data are all congruent in demonstrating persistent structural and functional changes to CNS structures and circuits including the prefrontal cortex, hippocampus, amygdala, and other cortical/subcortical areas of brain, with increasing evidence that the ELS-specific subtypes result in specific neuroanatomical alterations.

One study of children used structural magnetic resonance imaging (MRI) and executive function assessment do determine the relationship between ELS, executive function, and prefrontal cortex volume and connectivity with the anterior cingulate frontal poles. Increased ELS was associated with smaller PFC volumes in both gray and white matter between anterior cingulate and frontal poles, which was associated with poor executive functioning.[113]

The hippocampus has long been an area of interest for a multitude of reasons, one being that it is known to play a pivotal role in efficient termination of the HPA axis stress response by virtue of its rich density of GRs. Moreover, hippocampal volume reductions
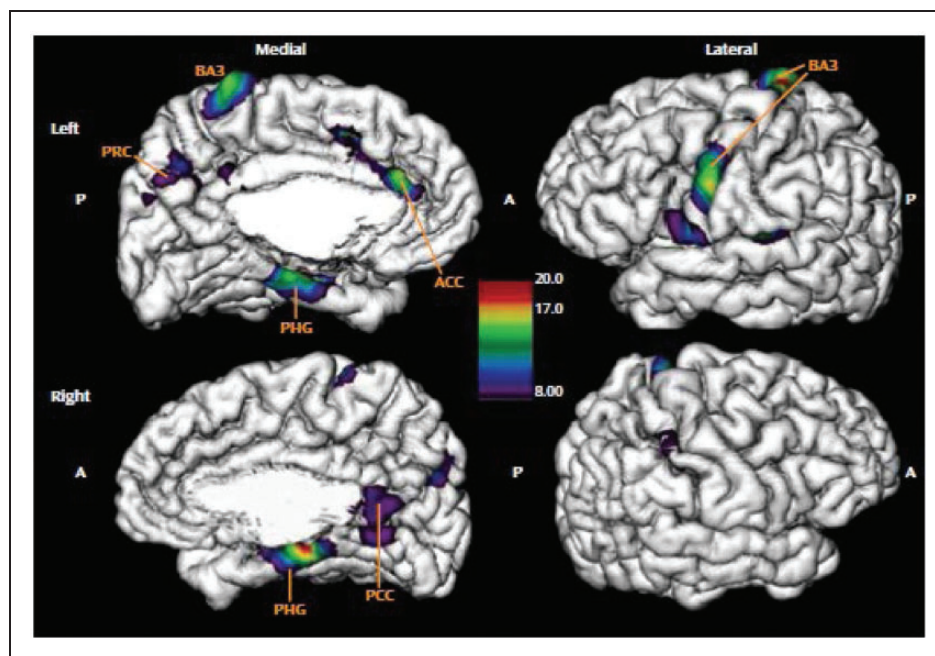
Figure 3. Regression of CTQ Total Score against cortical thickness in women with and without childhood sexual abuse. Cortical thickness analysis results after regressing CTQ total score against thickness across the entire cortex. Control variables included age and depression scores. Main effects are seen in the somatosensory cortex in the female genital and mouth area on the left, the PHG bilaterally, the left ACC, and the PRC bilaterally. For the precise location of the genital sensory field as identified using fMRI of neural response to stimulation, see Heim et al.[119] The color scale refers to the *F* values of the linear regression (significance threshold: $F > 4.33$). From Heim et al.[119] Reprinted with permission from the *American Journal of Psychiatry*.
BA3: Brodmann's area 3; PCC: posterior cingulate cortex; A: anterior; *p:* posterior; CTQ: Childhood Trauma Questionnaire; PHG = para-hippocampal gyrus; ACC = anterior cingulate cortex; PRC = precuneus; fMRI = functional magnetic resonance imaging.

have been repeatedly reported in those suffering from major depression, PTSD, and other psychiatric disorders. Reports of reduced hippocampal volume in depressed women with a history of childhood maltreatment but not in equally depressed women without ELS have also been confirmed by others.[114–116] This has been confirmed in a comprehensive meta-analysis (Figure 7).[117]

Another study compared depressed patients and age- and sex-matched healthy controls found that childhood maltreatment but not depression was associated with hippocampal atrophy (Figure 5).[118] Victims of childhood sexual and emotional abuse showed marked thinning in respective areas of cortical representation, suggesting that ELS has effect on neural plasticity that persist into adulthood (Figure 3).[119]

The pre-eminent role of the amygdala in stress responsivity has appropriately made it a central focus in research of mood and anxiety disorders. Both amygdala volume and responsiveness to stressors in those exposed to child abuse and neglect versus controls have been explored.

Non-human primates, subjected to ELS (variable foraging demand), demonstrated an increase in amygdala volume as assessed by MRI. This increase of amygdala volume was correlated with elevated CSF CRF concentrations, along with reduced hippocampal neurogenesis and increased anxiety.[120]

ELS also appears to alter connectivity between the amygdala and PFC, and there is a general consensus that depression is associated with increased amygdala responsiveness to stress. Whether this is a result of ELS or depression or a predisposing selective polymorphism still remains to be determined.

Threat-related amygdala reactivity was studied in adolescents, where it was shown to be positively associated with a family history of depression and severity of stressful life events.[121] This suggests that amygdala hyperactivity may precede manifestations of syndromal mood disturbance.

Childhood maltreatment (assessed by the Childhood Trauma Questionnaire) was shown to be positively associated with amygdala responsiveness in a standard emotional face-matching paradigm. This effect was not confounded by recent life stressors, current depression, or sociodemographic factors (Figure 6).[122]

Imaging genetic studies exploring some of the earlier discussed selective polymorphism in candidate genes
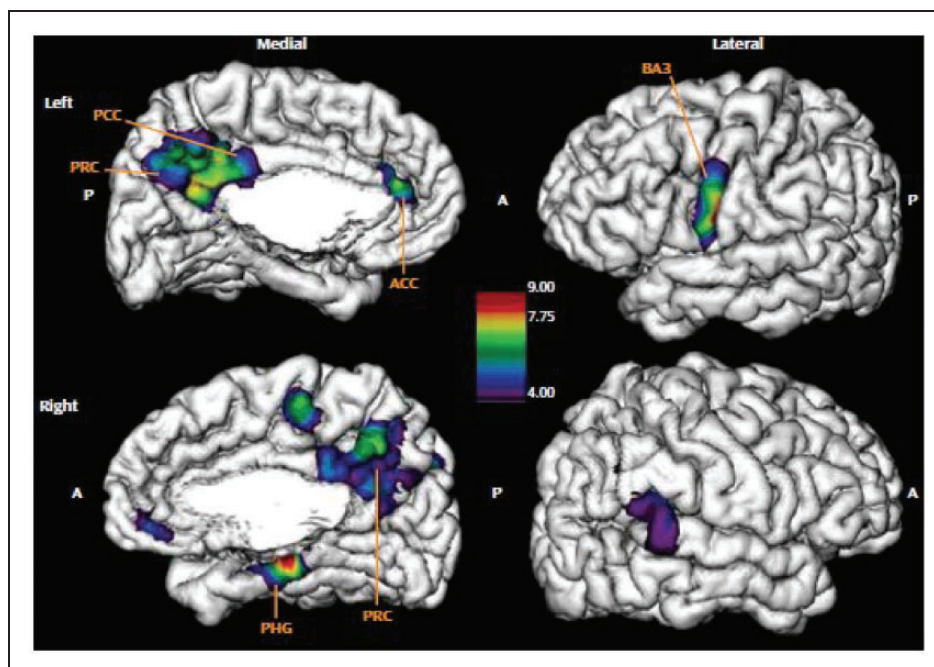
Figure 4. Regression of CTQ Emotional Abuse Score against Cortical Thickness in Women with and without Childhood Sexual Abuse. Cortical thickness analysis results after regressing CTQ emotional abuse score against thickness across the entire cortex. Control variables included age, depression, and all other CTQ subscales. Main effects are seen in the left and right PRC), left ACC, right PHG), and left somatosensory cortex in the area of the face. The color scale refers to the F values of the linear regression (significance threshold: $F > 4.33$). From Heim et al.[119] Reprinted with permission from the *American Journal of Psychiatry*.
BA3: Brodmann's area 3; PCC: posterior cingulate cortex; A: anterior; *p:* posterior; CTQ: Childhood Trauma Questionnaire; PHG = parahippocampal gyrus; ACC = anterior cingulate cortex; PRC = precuneus.
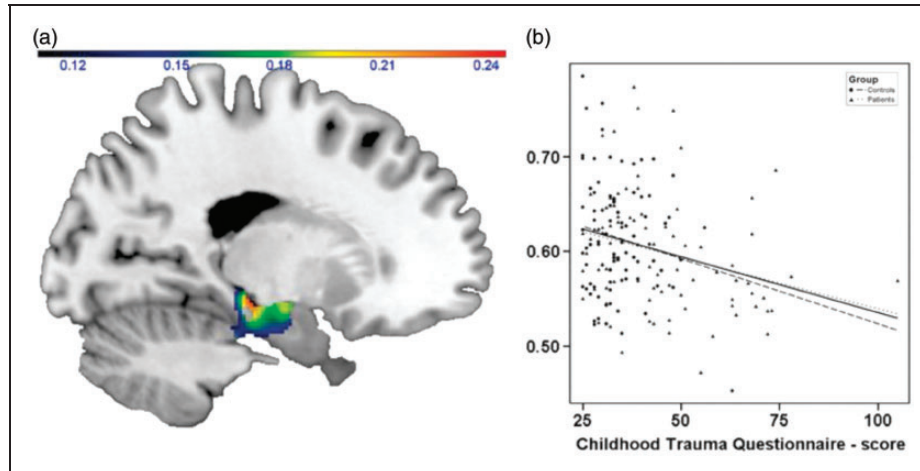
Figure 5. Effect of childhood maltreatment on hippocampal gray matter volume in the entire study sample. (a) Coronal view ($x = 0.75$, 14) depicting gray matter volume negatively associated with CTQscores; color bar, negative correlation coefficient r. (b) Scatter plot depicting gray matter volume at $x = 0.75$, 14; $y = 0.75$, 10; $z = 0.75$, 24 correlated with CTQ scores within the entire sample. Dotted lines: regression slopes of patients and controls separately; continuous line: regression slope in the entire sample. From Opel et al.[118] Reprinted by permission from Macmillan Publishers.
CTQ: Childhood Trauma Questionnaire.
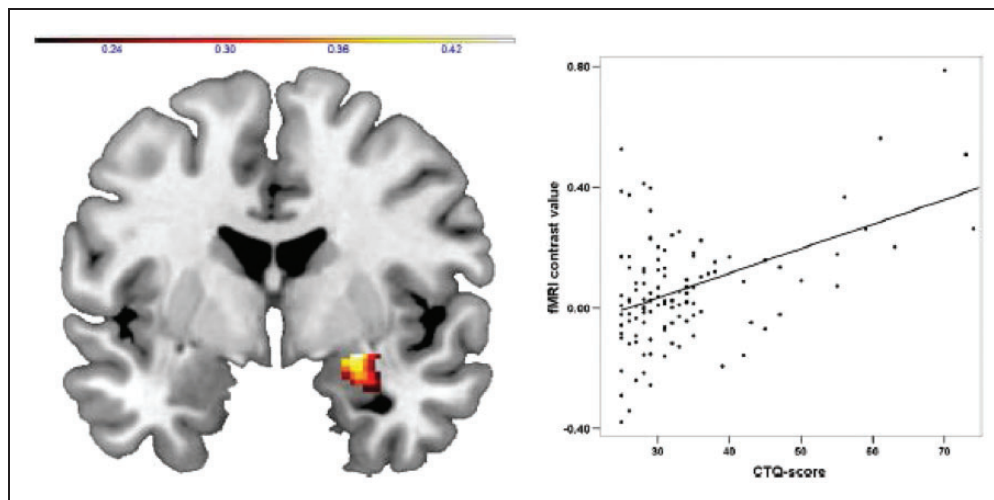
Figure 6. Childhood maltreatment, CTQ scores, is positively associated with right amygdala responsiveness to negative facial expressions. Left: coronal view ($y = !2$) depicting amygdala responsiveness modulated by CTQ scores. For display reasons, the statistical threshold was set to $p < 01$, uncorrected. Color bar, correlation coefficient r. Right: scatter plot depicting the positive correlation ($r = 0.456$, $p < .0001$) of the mean cluster activation values (left) and CTQ scores. From Dannlowski et al.[122] Reprinted with permission from Elsevier.
CTQ: Childhood Trauma Questionnaire.

have been found to correlate with altered amygdala response to stress. Evidence for FKBP5 and mineralocorticoid receptors modulating the effect of ELS on amygdala reactivity has been demonstrated.[123,124] Carriers of the *s* allele of the SERT have been found to have reduced gray matter volume in the amygdala as well as cingulate regions[125]; the *s* allele carrier also showed relative uncoupling of this circuit when processing fearful stimuli and was associated with temperamental anxiety.

How ELS Neurobiological Research Fits in the Era of RDoC

The emergence of the RDoC was in part due to the perceived failings of the current categorical DSM system to "capture fundamental underlying mechanisms of dysfunction" of psychiatric disorders.

The successful elucidation of pathophysiological mechanisms can be said to be the hallmark of much contemporary medicine and may be in part the reason why
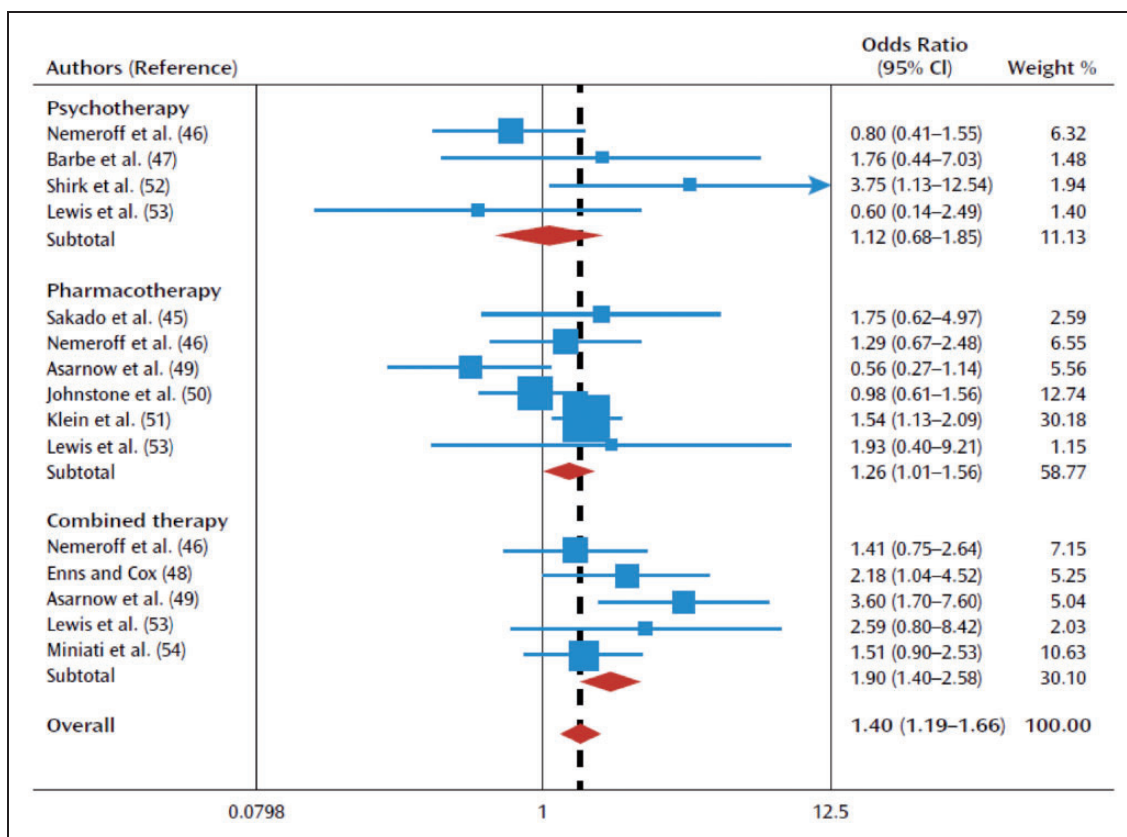
Figure 7. Meta-analysis of clinical trials investigating the association between childhood maltreatment and treatment outcome of depression. Based on the evidence of homogeneous distributions of effect sizes within treatment groups, we present here the results of fixed-effects model meta-analyses for different treatment groups. The overall effect size across treatment groups was estimated with a random-effects model meta-analysis with the following study weights: Nemeroff (psychotherapy): 7.88; Barbe: 2.78; Shirk: 3.49; Lewis (psychotherapy): 2.65; Sakado: 4.36; Nemeroff (pharmacotherapy): 8.03; Asarnow (pharmacotherapy): 7.32; Johnstone: 10.96; Klein: 14.09; Lewis (pharmacotherapy): 2.25; Nemeroff (combined therapy): 8.42; Enns: 7.07; Asarnow (combined therapy): 6.90; Lewis (combined therapy): 3.61; and Miniati: 10.18. The red diamonds show the combined effect sizes for studies concerned with psychotherapy, pharmacotherapy, and combined therapy as well as the overall effect size of the meta-analysis (top to bottom). From Nanni et al.[117] Reprinted with permission from the *American Journal of Psychiatry*.

advances in psychiatry have lagged behind other disciplines in medicine.[126]

One of the fundamental founding tenets of RDoC was that data from genetics and clinical neurosciences will reveal "bio-signatures" that will enhance the specificity of psychiatric diagnosis and treatment; in an age where precision medicine has resulted in important advances in translational research, genomics and neuroscience will likely pave the way for a framework that will allow psychiatric research to move forward in an accelerated manner.[127]

Because RDoC's primary focus is on neural circuitry in a bidirectional manner, ELS-neurobiological research findings to date as summarized above can be said to be a natural extension of the current domains and level of analysis, by incorporating the preeminent role of ELS in both preclinical and clinical research (environmental exposure) on development.

While more time and data are needed to discern the improvements RDoC will impart on the field of psychiatry, it may be said that it presents a paradigm shift in the field that fosters more emphasis on the underlying mechanisms mediating psychiatric disorders. This evolution of thought of psychiatric disorders may change the way in which psychiatrists of the future train and ultimately improve patient care.

The ELS paradigm, in both preclinical and clinical research, has generated a diverse aggregate of findings that range from $G \times E$ interactions to gross changes of hippocampal/amygdala volume and persistent perturbation of the HPA axis. ELS is clearly a major detriment to several of the currently defined diagnostic categories, including major depression, bipolar disorder, and PTSD.[128]

Indeed, as a framework that intends to integrate many different levels of data analysis within each domain/

construct, ELS can be said to play a role in almost every level as the scope of its effects encompass autonomic, immune, behavioral, endocrine, and circuit-level alterations that appear to interact with pathophysiological processes thought to potentially underlie these disorders.

Although psychiatry is considered to be one of the most "artful" areas of medicine, the field must develop biomarkers that will improve outcomes. This is one of the long-term goals of what RDoC hopes to achieve. ELS research has been able to begin to meet these demands via the core finding discussed in this review.

Declaration of Conflicting Interests

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