Differential functional connectivity within an emotion regulation neural network among individuals resilient and susceptible to the depressogenic effects of early life stress

J. M. Cisler[1]*, G. A. James[1], S. Tripathi[1], T. Mletzko[2], C. Heim[2], X. P. Hu[3], H. S. Mayberg[2], C. B. Nemeroff[4] and C. D. Kilts[1]

[1] *Brain Imaging Research Center, Psychiatric Research Institute, University of Arkansas for Medical Sciences, Little Rock, AR, USA*
[2] *Department of Psychiatry and Behavioral Sciences, Emory University, Atlanta, GA, USA*
[3] *Biomedical Imaging Technology Center, Emory University, Atlanta, GA, USA*
[4] *Department of Psychiatry and Behavioral Sciences, University of Miami Miller School of Medicine, Miami, FA, USA*

Background. Early life stress (ELS) is a significant risk factor for depression. The effects of ELS exposure on neural network organization have not been differentiated from the effect of depression. Furthermore, many individuals exposed to ELS do not develop depression, yet the network organization patterns differentiating resiliency *versus* susceptibility to the depressogenic effects of ELS are not clear.

Method. Women aged 18–44 years with either a history of ELS and no history of depression ($n=7$), a history of ELS and current or past depression ($n=19$), or a history of neither ELS nor depression ($n=12$) underwent a resting-state 3-T functional magnetic resonance imaging (fMRI) scan. An emotion regulation brain network consisting of 21 nodes was described using graph analyses and compared between groups.

Results. Group differences in network topology involved decreased global connectivity and hub-like properties for the right ventrolateral prefrontal cortex (vlPFC) and decreased local network connectivity for the dorsal anterior cingulate cortex (dACC) among resilient individuals. Decreased local connectivity and increased hub-like properties of the left amygdala, decreased hub-like properties of the dACC and decreased local connectivity of the left vlPFC were observed among susceptible individuals. Regression analyses suggested that the severity of ELS (measured by self-report) correlated negatively with global connectivity and hub-like qualities for the left dorsolateral PFC (dlPFC).

Conclusions. These preliminary results suggest functional neural connectivity patterns specific to ELS exposure and resiliency *versus* susceptibility to the depressogenic effects of ELS exposure.

Introduction

Early life stress (ELS) refers to an array of major adversities occurring before sexual maturation, including physical, sexual and emotional abuse, physical and emotional neglect, malnourishment, and loss of a parent (Heim *et al.* 2000, 2004; Gillespie & Nemeroff, 2007). ELS is a significant risk factor for the development of a broad array of mental health disorders, including major depressive disorder (MDD) (Resnick *et al.* 1993; Kessler *et al.* 1997; McCauley *et al.* 1997; Kilpatrick *et al.* 2003). Numerous neurobiological consequences of ELS may contribute to the heightened risk for MDD (Heim *et al.* 2004; Gillespie & Nemeroff, 2007). Human studies have demonstrated an association of ELS with persistent sensitization of the neuroendocrine, autonomic and neuroimmune responses to stress (Heim *et al.* 2000; Carpenter *et al.* 2010). Human neuroimaging research suggests neuroanatomical alterations following ELS, including decreased hippocampal, orbitofrontal cortex (OFC) and dorsolateral prefrontal cortex (dlPFC) volumes (Vythilingam *et al.* 2002; Hanson *et al.* 2010), in addition to functional alterations, including altered activation of the amygdala, posterior insula and dorsal anterior cingulate cortex (dACC) during cognitive control and emotion processing tasks (Taylor *et al.* 2006; Weber *et al.* 2009; Williams *et al.* 2009; Matz *et al.* 2010; Mueller *et al.* 2010).

* Address for correspondence : J. M. Cisler, Ph.D., Brain Imaging Research Center, Psychiatric Research Institute, University of Arkansas for Medical Sciences, 4301 West Markham St, Mail Slot 554, Little Rock, AR 72205, USA.

(Email : jcisler@uams.edu)

This consistent evidence for altered neural processing within brain networks mediating emotion processing and emotion regulation among individuals with ELS histories suggests how ELS may increase risk for affective disorders such as depression (although it should be noted that depression is not necessarily always preceded by ELS). Individuals with a history of depression demonstrate smaller hippocampal volumes (Bremner *et al.* 2000*a*; Vythilingam *et al.* 2002) and altered activation of the amygdala, subgenual and rostral ACC, hippocampus, caudate, insula and PFC during cognitive control and emotion processing tasks independent of a history of ELS (Davidson *et al.* 2003; Anand *et al.* 2005; Johnstone *et al.* 2007; Lee *et al.* 2008; Matthews *et al.* 2008; Victor *et al.* 2010; Elliott *et al.* 2011). Furthermore, path and connectivity analysis models of depression also demonstrate altered networks mediating emotion perception and regulation (Seminowicz *et al.* 2004; Greicius *et al.* 2007; Matthews *et al.* 2008; James *et al.* 2009; Vasic *et al.* 2009; Erk *et al.* 2010). Given the neurobiological overlap between ELS and depression, it is important to further characterize the specific neural network organization patterns associated with ELS and depression to better separate risk factors from markers of psychopathology.

Although overlapping dysfunction in neural networks mediating emotion regulation for ELS and depression suggests how ELS might confer vulnerability to depression, it does not inform observed individual variation in depression vulnerability associated with ELS. The modes of neural information processing that mediate susceptibility and resiliency to MDD following ELS are not known. Elucidation of the specific network organization patterns of individuals resilient to the effects of ELS would provide an understanding of how the brain mediates healthy recovery from ELS, which would inform methods of prevention of ELS-related psychopathology and also provide viable neural processing targets for the treatment of ELS-related psychopathology.

The purpose of the current study was to provide an exploratory examination of specific emotion regulation network organization effects of (1) ELS exposure *per se*, (2) resiliency to the effects of ELS and (3) susceptibility to the effects of ELS. Accordingly, we used graph theoretical analyses to compare organization of a 21-node emotion regulation network between a control group with a history of neither ELS nor depression, a resilient group of individuals with a history of ELS but no history of depression (or other psychopathology), and a susceptible group of individuals with a history of both ELS and depression. Functional connectivity (FC) measured during rest is independent of task demands and is thought to represent intrinsic FC pathways (Deco *et al.* 2011). Indeed, FC identified during rest corresponds well with underlying structural pathways (van den Heuvel *et al.* 2009). Accordingly, use of resting-state data allows an examination of intrinsic FC pathways among resilient and susceptible individuals.

Method

Participants

Participants included 19 women with a history of ELS and either past or current DSM-IV-defined MDD (i.e. 'susceptible' individuals), seven women with a history of ELS but no diagnosis of current or past MDD (i.e. 'resilient' individuals), and 12 women with a history of neither ELS nor MDD (i.e. 'controls'). Participants were recruited through advertisements in local newspapers and on public transport, and also through information flyers posted throughout the Metro Atlanta Area in highly frequented places such as coffee shops and grocery stores. Table 1 summarizes the demographic and clinical characteristics of the three groups. The participants with current and past MDD were combined into one group because the hypothesis tested in the current study centers around resiliency *versus* susceptibility. Because participants with current and past MDD both demonstrate susceptibility to depression following ELS, they were combined into one group.

ELS and MDD histories were collected with the Structured Interview for Clinical Disorders (SCID; First *et al.* 2002) and the Early Trauma Inventory (ETI; Bremner *et al.* 2000*b*). All participants additionally completed the Childhood Trauma Questionnaire (CTQ; Bernstein *et al.* 1994) and the Hamilton Depression Rating Scale (HAMD; Hamilton, 1960) to further characterize their ELS history and current MDD symptoms respectively. Current or past DSM-IV diagnoses of MDD were determined based on the SCID, and ELS history was determined by the ETI and CTQ. Exclusion criteria were significant medical illness (e.g. gastrointestinal, neurological, endocrine, cardiovascular disorders), pregnancy or nursing, past or current presence of psychotic symptoms or bipolar disorder, current presence of psychoactive substance abuse/dependency or eating disorders, hormonal medication (except for oral contraceptives), and psychotropic medication in the 2 weeks prior to study entry (4 weeks for selective serotonin reuptake inhibitors, SSRIs). For subjects assigned to the non-ELS groups, exclusion criteria were major stress experiences before the onset of puberty, such as abuse, neglect, separation from parents, parental death, accidents, severe illness or natural disaster. Participants were compensated for their time in the study.

Table 1. *Demographic and clinical characteristics of the sample*

	Control ($n = 12$)	ELS no MDD ($n = 7$)	ELS with MDD ($n = 19$)
Age (years), mean (S.D.)	25.92 (5.33)	27.43 (7.39)	31.28 (8.57)
Race			
% Caucasian	46	43	17
% Black	23	29	67
% Asian	23	14	6
% Hispanic	8	14	6
CTQ total score, mean (S.D.)	27.25 (1.87)$_{a,b}$	42.71 (15.00)$_{b,c}$	66.63 (18.50)$_{a,c}$
CTQ emotional abuse, mean (S.D.)	5.50 (0.80)$_{a,b}$	11.71 (7.06)$_{b,c}$	16.26 (5.23)$_{a,c}$
CTQ physical abuse, mean (S.D.)	5.75 (1.06)$_a$	7.71 (2.93)	11.05 (5.10)$_a$
CTQ sexual abuse, mean (S.D.)	5.00 (0.00)$_a$	5.14 (0.38)$_b$	14.05 (8.61)$_{a,b}$
CTQ emotional neglect, mean (S.D.)	6.00 (1.35)$_{a,b}$	11.29 (5.25)$_{b,c}$	15.42 (4.39)$_{a,c}$
CTQ physical neglect, mean (S.D.)	5.00 (0.00)$_a$	6.86 (3.19)$_b$	9.84 (3.75)$_{a,b}$
HAMD, mean (S.D.)	1.50 (0.80)$_a$	2.43 (1.72)$_b$	11.42 (8.93)$_{a,b}$
Current MDD, n (%)	0 (0)	0 (0)	6 (33)
Lifetime MDD, n (%)	0 (0)	0 (0)	19 (100)
Current PTSD, n (%)	0 (0)	0 (0)	7 (37)
Lifetime PTSD, n (%)	0 (0)	0 (0)	10 (53)

ELS, Early life stress; MDD, major depressive disorder; CTQ, Childhood Trauma Questionnaire; HAMD, Hamilton Depression Rating Scale; PTSD, post-traumatic stress disorder; S.D., standard deviation.

Across the columns, data points that share a common subscript indicate that those groups differ significantly on that variable ($p < 0.05$).

MRI acquisition

Neuroimaging data were acquired on a 3.0-T Siemens Magnetom Trio scanner (Siemens Medical Solutions USA) with a Siemens transmit–receive head coil. Anatomical images were acquired at $1 \times 1 \times 1$ mm³ resolution with a magnetization prepared rapid gradient echo (MPRAGE) sequence as 176 1-mm-thick sagittal slices with the following parameters: field of view (FOV) = 224×256 mm, repetition time (TR) = 2600 ms, echo time (TE) = 3.02 ms, and flip angle (FA) = $8°$. Functional images were acquired with a *z*-saga sequence (Heberlein & Hu, 2004) to minimize artifacts in the medial PFC (mPFC) and OFC due to sinus cavities. Z-saga images were acquired at $3.4 \times 3.4 \times 4$ mm resolution in 20 4-mm-thick axial slices with the following parameters: FOV = 220×200 mm, TR = 2020 ms, $TE^1/TE^2 = 30$ ms/66 ms, FA = $90°$ for 210 acquisitions, and total duration = 7.2 min. During the resting-state scan, participants were instructed to lie passively in the scanner and to refrain from thinking about anything specific.

Image preprocessing

Image preprocessing followed standard steps and was completed using AFNI software (NIMH, USA). In order, images underwent slice timing correction, motion correction, alignment to participant's normalized anatomical images, despiking, transformation into percentage signal change, detrending, and were passed through a low-frequency bandpass filter (0.009–0.08 Hz) and spatially smoothed using a 6 mm full-width at half-maximum (FWHM) Gaussian filter. Images were normalized using the Montreal Neurological Institute (MNI) 452 template brain. In addition, to correct for artifacts due to the sequential slice acquisition sequence used by the *z*-saga routine, fluctuations in white matter voxels and the cerebrospinal fluid (CSF) were regressed out of time courses from gray matter voxels separately for each axial slice following segmentation using FSL (Franco *et al.* 2011), and this step was implemented directly after detrending in the preprocessing stream.

Emotion processing and emotion regulation network node selection and extraction

Based on brain anatomical coordinates provided by neuroimaging studies of emotion processing and emotion regulation in healthy and depressed individuals, and from network models of depression (Craig, 2002; Ochsner *et al.* 2002, 2004; Critchley *et al.* 2004; Seminowicz *et al.* 2004; Greicius *et al.* 2007; Johnstone *et al.* 2007; Matthews *et al.* 2008; Wager *et al.* 2008; Craddock *et al.* 2009; James *et al.* 2009; Erk *et al.* 2010; McRae *et al.* 2010; Price & Drevets, 2010; Elliott *et al.* 2011), we stipulated an emotion processing and regulation network consisting of

Table 2. *Name (abbreviations) and MNI coordinates of centroids of nodes comprising an affective cognition network*

Region of interest (abbreviation)	MNI coordinates		
	x	y	z
Left hippocampus (lHPC)	−28	−22	−12
Right hippocampus (rHPC)	28	−22	−12
Right amygdala (rAMY)	22	−2	−15
Left amygdala (lAMY)	−20	−4	−15
Right dorsolateral PFC (rDFC)	32	42	32
Left dorsal lateral PFC (lDFC)	−32	42	32
Right thalamus (rT)	10	−14	10
Left thalamus (lT)	−10	−14	10
Left caudate (lC)	−14	12	12
Right caudate (rC)	14	12	12
Subgenual cingulate (sACC)	1	25	−11
Rostral anterior cingulate (rACC)	2	36	6
Dorsal anterior cingulate (dACC)	1	14	30
Medial PFC (mFC)	1	60	6
Ventral medial PFC (vmFC)	1	51	−9
Left ventral lateral PFC (lvFC)	−44	38	−8
Right ventral lateral PFC (rvFC)	44	38	−8
Right anterior insula (raI)	38	20	0
Left anterior insula (laI)	−36	20	0
Right posterior insula (rpI)	42	−12	2
Left posterior insula (lpI)	−42	−12	2

21 nodes (see Table 2 and Fig. 1). Anatomical co-ordinates of the nodes used in the present study were guided by coordinates provided in these studies, such that the centroid of the coordinates was shifted as needed, so that a 6-mm sphere applied to the International Consortium for Brain Mapping (ICBM) 452 template was located in gray matter and co-ordinates in Talaraich space were first converted to MNI space. A trained neuroscientist (G.A.J.) confirmed the accuracy of the node's placement with respect to its intended location. Time courses were extracted from 6-mm-radius spherical regions of interest (ROIs) centered at the centroid coordinates of each node for each individual and averaged across voxels within an ROI, resulting in a 21 × 210 (node by time point) matrix for each individual.

Graph theoretical analyses

Graph analyses examine complex network organizations by representing a network as a graph consisting of nodes (or vertices) and edges (or connections between nodes) (Bullmore & Sporns, 2009; Bullmore & Bassett, 2011; Rubinov & Sporns, 2010). For binary undirected graphs (i.e. an edge either exists or does not and causality between nodes is not implied), the graph's nodes are represented by ROIs and its edges are represented by the correlation matrix of the ROIs' time series. A threshold T is applied to the correlation matrix of the ROI time series, such that $r \geqslant T$ is considered an edge connecting the two nodes and $r < T$ is not considered an edge. Undirected (i.e. no causal direction implied) graphs were created separately for each individual using the following steps. First, a correlation matrix was created by correlating the time courses of each ROI, resulting in a 21 × 21 correlation matrix. Second, consistent with prior research (Bullmore & Bassett, 2010; Lynall *et al.* 2010; van Wijk *et al.* 2010), edges were defined using a range of threshold values based on connection density (i.e. ratio of the number of connections in a network to the total number of connections possible). Simulation studies demonstrate that graph network indices differ as a function of density (van Wijk *et al.* 2010), thus it is necessary to define edges using a thresholding procedure that controls for connection density differences across individuals and groups. We considered only connected graphs (graphs where no node is unconnected from the rest of the graph) and defined T at a range of densities from 0.37 to 0.50 in 0.01 increments (Lynall *et al.* 2010). This range of densities was chosen because densities <0.37 tend to be fragmented and densities >0.50 may not exist in biological systems (Lynall *et al.* 2010). Following identification of each threshold value, a 21 × 21 binary adjacency matrix was calculated from the correlation matrix, in which a correlation $\geqslant T$ was coded 1, a correlation $< T$ was coded 0, and the diagonals were all set to 0. This resulted in 14 separate 21 × 21 binary adjacency matrices for each individual (representing densities ranging from 0.37 to 0.50). Third, network indices (detailed in the following sections) were calculated using Matlab toolboxes (provided by Rubinov & Sporns, 2010) for each of these adjacency matrices and then averaged together (Lynall *et al.* 2010).

Graph network indices

Degree (k_i)

k_i is the number of connections (edges) linking node i to the rest of the graph. For example, a node connected to four other nodes has a degree of 4. This index gives a measure of how functionally connected a node is to the overall network.

Efficiency (E_i)

E_i is the inverse of the mean path length connecting node i to each other node in the network. Path length represents the number of links needed to connect two nodes (e.g. a direct connection between nodes i and j represents a path length of 1; an indirect
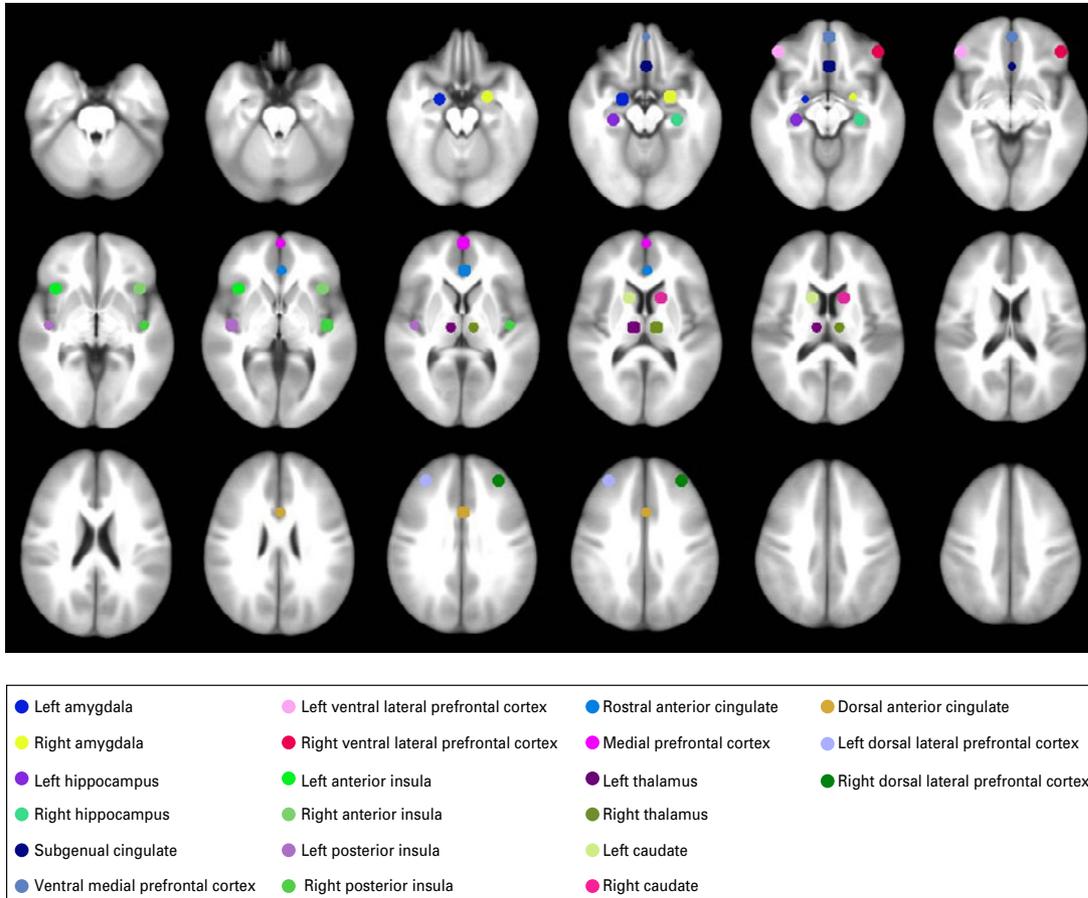
Fig. 1. Regions of interest (ROIs) overlaid on top of the International Consortium for Brain Mapping (ICBM) 452 template brain.

connection of nodes *i* and *j* through node *k* represents a path length of 2), and mean path length of node *i* represents the mean of all path lengths connecting node *i* to every other node. Efficiency is considered a more robust network statistic compared to path length (Rubinov & Sporns, 2010) because path length is disproportionately affected by unconnected nodes (i.e. disconnected nodes have infinitely long path lengths). By contrast, a disconnected node has an efficiency of 0; thus, disconnected nodes do not have a disproportionate effect on efficiency. This metric gives an indication of how efficiently information within one node can get to other nodes in the network.

Clustering coefficient (C_i)

C_i is the ratio of a node *i*'s neighbors that are also neighbors of each other. One example might be a situation in which node *i* is connected to nodes *j*, *k* and *l*, and only nodes *j* and *k* are also connected. This metric provides a measure of local 'cliques', where information processing/FC is more segregated from the rest of the network.

Betweenness centrality (B_i)

B_i refers to the fraction of all shortest paths in a network that pass through node *i*. Nodes with high B_i are considered 'hubs,' such that these nodes tend to link more segregated nodes or regions of nodes. Because they operate as 'hubs' linking the network together, deletion of nodes with high between centrality has a detrimental effect on a network's functioning (Albert *et al.* 2000).

Analyses

Assessment of group effects on network connectivity proceeded in three steps. First, omnibus group contrasts for each of these specific effects were defined, with the specific effect of ELS exposure defined by the contrast of the two ELS-exposed groups ($n = 26$) *versus* the control group ($n = 12$), the specific effect of resilience to ELS defined by the contrast of the resilient group ($n = 7$) *versus* the combination of the control and susceptible groups ($n = 31$), and the specific effect of susceptibility to ELS defined by the contrast of the susceptible group ($n = 18$) to the combination of the

resilient and control groups ($n=20$). These contrasts were conducted for each of the four network indices for each of the 21 nodes. To correct for multiple comparisons, the p value of each contrast was adjusted to a family-wise corrected α of 0.05 using Bootstrap permutation testing implemented in SAS 9.2 PROC MULTTEST (SAS Institute Inc., USA). Second, following the identification of significant group effects from the omnibus contrasts, follow-up tests compared the control, resilient and susceptible groups to confirm the specific network effects from the pooled group contrasts. Significant effects emerging from both of these procedures were then plotted to visualize the results. Third, linear multiple regression analyses were used to examine scalar relationships between each node's network statistics and the severity of ELS (measured by the CTQ total score) and of MDD symptoms (measured with the HAMD). These regressions entered age, race (dichotomized to Caucasian *versus* minority), centered CTQ total score, centered HAMD score, and the CTQ × HAMD interaction term. These analyses allow for an examination of the specific effect of ELS and MDD symptoms on network organization (i.e. the effect of ELS when controlling for MDD and vice versa). The raw p values for ELS and MDD from each regression model were then adjusted using the false discovery rate method implemented in SAS 9.2 PROC MULTTEST.

Given that this is an exploratory graph theoretical analysis applied to the effects of ELS exposure and resiliency and susceptibility to the depressogenic effects of ELS, we report both the corrected and uncorrected p values. Reporting of the effects that survive both corrected and uncorrected statistical thresholds will facilitate power analyses for the design of future studies.

Results

Differences in node network statistics between groups

Degree

For the emotion processing and emotion regulation network, the omnibus resiliency group contrast demonstrated decreased degree for the right ventrolateral PFC (vlPFC) ($p_{uncorr}=0.012$, $p_{corr}=0.19$) nodes. Follow-up group comparisons demonstrated decreased degree for the right vlPFC node compared to both the susceptible ($p=0.027$) and control ($p=0.015$) groups (Fig. 2*a*). The omnibus susceptibility contrast demonstrated increased degree of the left caudate among the susceptible group ($p_{uncorr}=0.038$, $p_{corr}=0.49$), and follow-up tests demonstrated increased degree among the susceptible group compared to the control

group ($p=0.038$) (Fig. 2*b*). The omnibus ELS contrast demonstrated increased degree of the right hippocampus ($p_{uncorr}=0.039$, $p_{corr}=0.51$), and follow-up tests demonstrated greater degree among the resilient group compared to the control group ($p=0.02$) (Fig. 2*c*).

Clustering coefficient

The omnibus resilience group contrast demonstrated decreased clustering for the dACC node ($p_{uncorr}=0.011$, $p_{corr}=0.14$) and increased clustering for the right vlPFC node ($p_{uncorr}=0.044$, $p_{corr}=0.44$). Follow-up group comparisons demonstrated less clustering for the dACC node in the resilient group compared to both the susceptible ($p=0.014$) and control ($p=0.033$) groups (Fig. 2*d*). There was increased clustering for the right vlPFC (Fig. 2*e*) in the resilient group compared to the control group ($p=0.042$) and marginally greater clustering compared to the susceptible group ($p=0.087$). The omnibus susceptibility and ELS group contrasts did not reveal any significant differences.

Efficiency

The omnibus resiliency group contrast demonstrated decreased efficiency for the dACC node ($p_{uncorr}=0.017$, $p_{corr}=0.21$) among the resilient group. Follow-up group comparisons demonstrated decreased efficiency for the dACC node compared to both the susceptible ($p=0.017$) and control ($p=0.013$) groups (Fig. 2*f*). The omnibus susceptibility group contrast demonstrated decreased efficiency for the left amygdala ($p_{uncorr}=0.026$, $p_{corr}=0.14$) and left vlPFC ($p_{uncorr}=0.014$, $p_{corr}=0.18$). Follow-up contrasts demonstrated lower efficiency for the amygdala compared to the control ($p=0.046$) but not the resilient group ($p=0.12$) (Fig. 2*g*), and lower efficiency for the left vlPFC compared to both the control ($p=0.03$) and the resilient ($p=0.015$) groups (Fig. 2*h*). The omnibus ELS contrast demonstrated increased efficiency of the mPFC among the ELS groups ($p_{uncorr}=0.035$, $p_{corr}=0.18$), and follow-up tests demonstrated increased efficiency among the resilient group compared to the control group ($p=0.025$) and marginally increased efficiency among the susceptible group relative to the control group ($p_{uncorr}=0.09$) (Fig. 2*i*).

Betweenness centrality

The omnibus resiliency group contrast demonstrated decreased betweenness centrality for the right vlPFC node ($p_{uncorr}=0.0048$, $p_{corr}=0.08$). Follow-up group comparisons demonstrated decreased centrality for the right vlPFC compared to both the susceptible
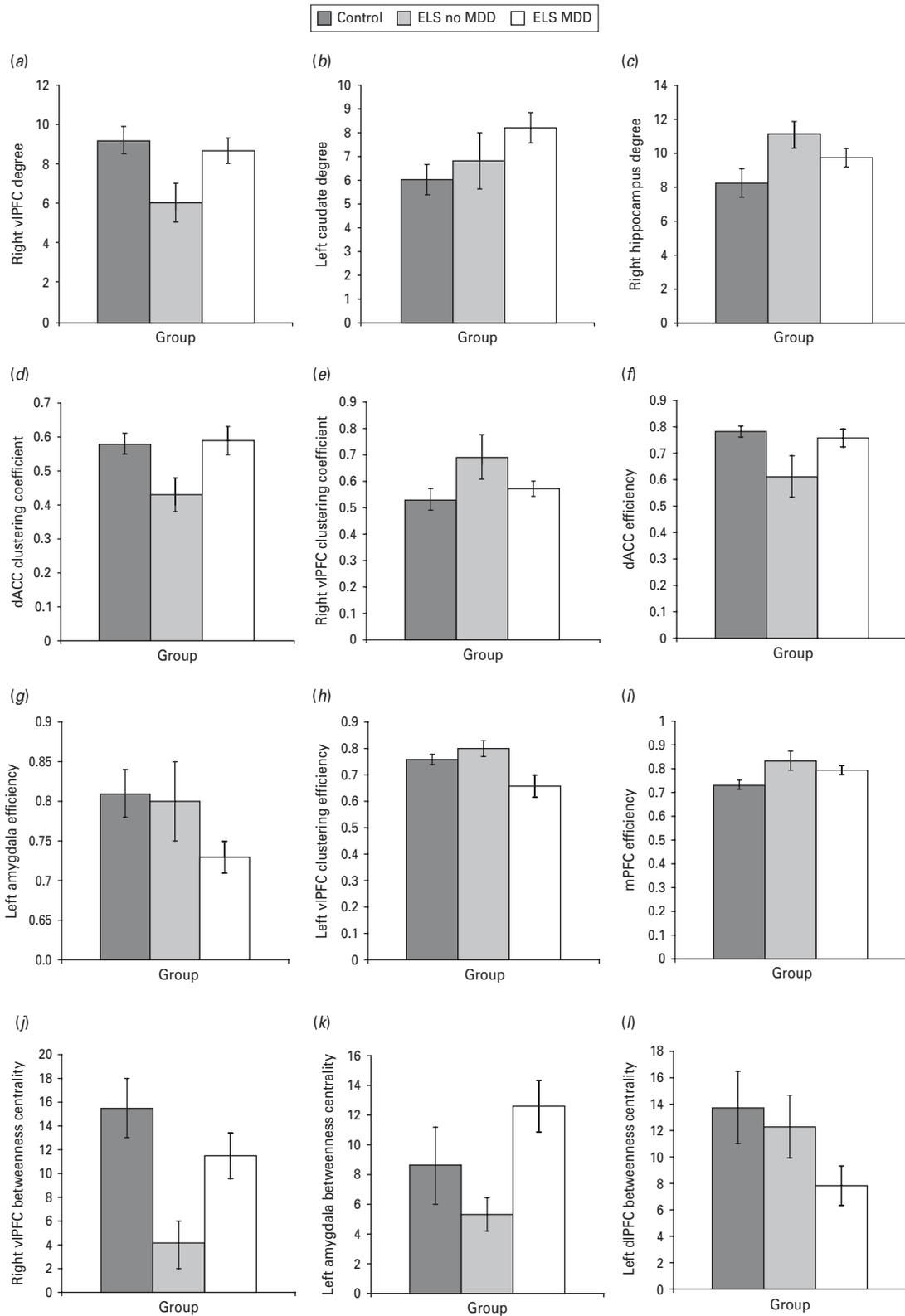
Fig. 2. Mean node network statistics (and error bars) demonstrating between-group differences: (*a*) right ventrolateral prefrontal cortex (vlPFC) degree, (*b*) left caudate degree, (*c*) right hippocampus degree, (*d*) dACC clustering coefficient, (*e*) right vlPFC clustering coefficient, (*f*) dACC efficiency, (*g*) left amygdala efficiency, (*h*) left vlPFC clustering efficiency, (*i*) medial PFC (mPFC) efficiency, (*j*) right vlPFC betweenness centrality, (*k*) left amygdala betweenness centrality, (*l*) dorsolateral PFC (dlPFC) betweenness centrality. ELS, Early life stress; MDD, major depressive disorder.

Table 3. *Summary of network statistic differences across groups*

Group	Node (ROI)	Finding
Resilient	Right vlPFC	Decreased degree relative to susceptible ($p=0.027$)
		Decreased degree relative to control ($p=0.015$)
		Increased clustering relative to susceptible ($p=0.087$)
		Increased clustering relative to control ($p=0.042$)
		Decreased betweenness centrality relative to susceptible ($p=0.038$)
		Decreased betweenness centrality relative to control ($p=0.004$)
	mPFC	Increased efficiency relative to control ($p=0.025$)
	Right hippocampus	Increased degree relative to control ($p=0.02$)
	dACC	Decreased clustering relative to susceptible ($p=0.014$)
		Decreased clustering relative to control ($p=0.033$)
		Decreased efficiency relative to susceptible ($p=0.017$)
		Decreased efficiency relative to control ($p=0.013$)
Susceptible	Left amygdala	Decreased efficiency relative to control ($p=0.046$)
		Increased betweenness centrality relative to resilient ($p=0.031$)
	Left vlPFC	Decreased efficiency relative to resilient ($p=0.015$)
		Decreased efficiency relative to control ($p=0.03$)
	Left dlPFC	Decreased betweenness centrality relative to control ($p=0.039$)
	Left caudate	Increased degree relative to control ($p=0.038$)

ROI, Region of interest; vlPFC, ventrolateral prefrontal cortex; dACC, dorsal anterior cingulate cortex; mPFC, medial PFC; dlPFC, dorsolateral PFC.

($p=0.038$) and control ($p=0.004$) groups (Fig. 2*j*). The omnibus susceptibility group contrast demonstrated increased betweenness centrality among the left amygdala ($p_{\text{uncorr}}=0.011$, $p_{\text{corr}}=0.19$) and decreased betweenness centrality of the left dlPFC ($p_{\text{uncorr}}=0.035$, $p_{\text{corr}}=0.46$). Follow-up tests demonstrated increased amygdala betweenness centrality among the susceptible group compared to the resilient group ($p=0.031$) (Fig. 2*k*), and less betweenness centrality of the dlPFC among the susceptible group compared to the control group ($p=0.039$) (Fig. 2*l*). Table 3 provides a summary of all comparisons.

Scalar relationships between node network statistics and ELS severity and MDD symptoms

The linear multiple regression analyses identified two specific predictors of network organization: ELS severity was significantly negatively correlated with the network indices of degree (partial $r=-0.499$, $p_{\text{uncorr}}=0.002$, $p_{\text{corr}}=0.048$) and betweenness centrality (partial $r=-0.533$, $p_{\text{uncorr}}=0.001$, $p_{\text{corr}}=0.021$) of the left dlPFC (Fig. 3). MDD severity was not significantly related to any network index while controlling for the other variables in the model.

Discussion

It should first be emphasized that this was an exploratory study and the results must be considered preliminary. Indeed, we report both corrected and uncorrected p values to emphasize the nature of the experimental effects with respect to corrections for multiple corrections. The strongest group contrast findings from this exploratory study suggest altered integration of the right and left vlPFC, dACC, left dlPFC and left amygdala into an emotion processing and emotion regulation network among resilient *versus* susceptible individuals. There is modest evidence of neural organizational consequences of ELS exposure *per se*, including greater clustering of the mPFC and greater degree of the right hippocampus. Finally, correlation analyses demonstrated that ELS severity, but not MDD symptoms, correlated negatively with left dlPFC degree and betweenness centrality measures of the network topology.

These preliminary results provide novel insights into the neural organization of ELS exposure and resiliency *versus* susceptibility to depression following ELS during the resting state. Specifically, the largest group differences tended to be between individuals resilient *versus* susceptible to depression following ELS. When examining the overall topological characteristics of the network, the preliminary results seem to suggest a more distributed pattern of information processing in this network among resilient individuals. Consistent with this notion, there was less integration (i.e. lower degree, betweenness centrality, efficiency) of the right vlPFC and dACC into the network, suggesting that function typically localized
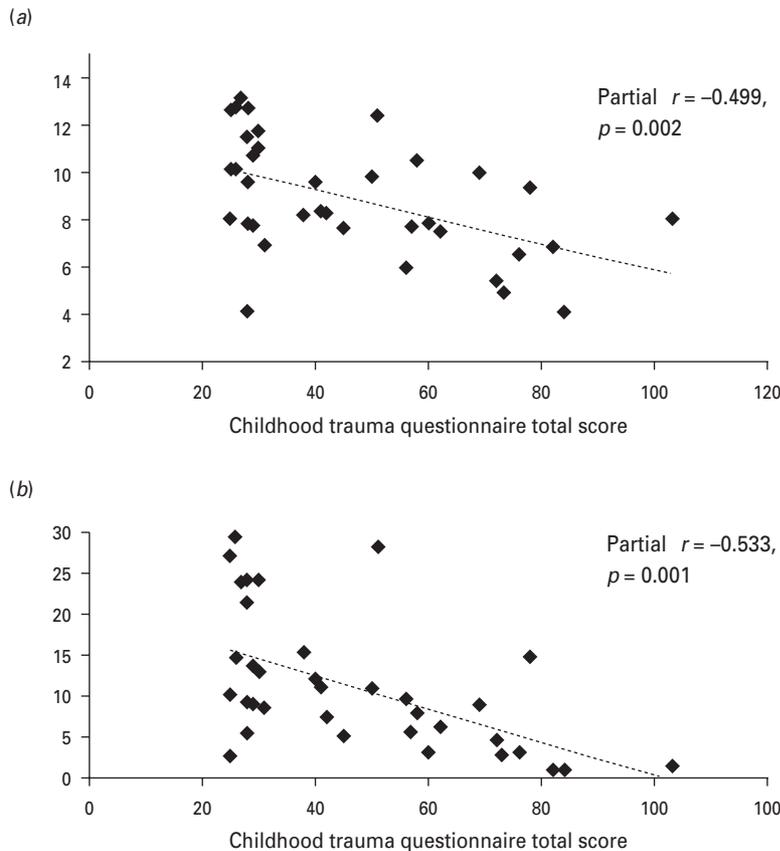
(*a*)

(*b*)

Fig. 3. Relationships between early life stress (ELS) severity [Childhood Trauma Questionnaire (CTQ) total score] and (*a*) left dorsolateral prefrontal cortex (dlPFC) degree and (*b*) left dlPFC betweenness centrality. The partial *r* values come from multiple linear regression analyses in which age, minority status, depression symptoms [Hamilton Depression Rating Scale (HAMD) total score] and the CTQ × HAMD interaction were also entered as predictors. ◆, dlPFC.

within these regions was instead distributed across the network. Simulation research has found that deletion of nodes with high betweenness centrality has a greater detrimental effect on network function relative to deletion of other nodes (Albert *et al.* 2000; Alstott *et al.* 2009); thus, a network is more stable if its information processing is distributed across many nodes instead of being reliant on fewer 'hub' nodes. Accordingly, the generally lower integration among nodes within this network among resilient individuals provides preliminary evidence of a more stable network.

Preliminary evidence was found that degree and betweenness centrality of the right vlPFC was reduced among resilient individuals, although clustering of this region was higher among resilient individuals. The clustering coefficient provides an index of local 'cliques', and this finding suggests that whereas the right vlPFC was generally less integrated into the network as a whole, the vlPFC and its neighborhood retained greater local connectivity. This pattern of reduced global integration with higher local integration may indicate that the right vlPFC and its neighbors are

more network independent and segregated. Given the importance of the vlPFC in engaging emotion regulation and inhibitory processes (Aron *et al.* 2004, 2007; Aron & Poldrack, 2006; Wager *et al.* 2008), it is tempting to hypothesize that greater segregation of this region's local neighborhood optimizes performance by reducing conflicting demands on the local neighborhood. However, this hypothesis remains speculative in the absence of task data to corroborate a correlation between segregation of right vlPFC's local neighborhood and performance.

We also present preliminary evidence of increased betweenness centrality, but lower efficiency, for the left amygdala among the susceptible group. The amygdala is a neural region crucial for detecting and processing emotionally and motivationally salient cues (LeDoux, 2000; Davis & Whalen, 2001) and is widely conceptualized as being over-reactive in models of depression (Price & Drevets, 2010; Elliott *et al.* 2011), which may explain heightened negative affect and rumination among depressed individuals. The lowered efficiency, yet higher betweenness centrality,

may help to explain altered functional activations of this node among depressed individuals. Lowered efficiency for this node suggests lowered local connectivity patterns (i.e. information processing has to transverse more neural regions to influence processing in the left amygdala), which suggests greater segregation of the left amygdala. As might be the case with the more segregated vlPFC among the resilient group, the more segregated left amygdala in the susceptible group may optimize functions performed by the amygdala (e.g. detecting emotional salient cues) by reducing conflicting/modulating inputs. However, whereas optimized inhibitory performance mediated by the vlPFC among resilient individuals seems to be an adaptive mechanism, enhanced emotion processing mediated by the amygdala in the susceptible group may be a maladaptive mechanism. Similarly, the greater betweenness centrality suggests that the shortest path lengths between other nodes in the network generally transverse the left amygdala, which suggests that the left amygdala in susceptible individuals may play a larger role in generally biasing information processing throughout the whole emotion processing and emotion regulation network. The preliminary finding of amygdala processing biasing information processing throughout the network seems to be consistent with a maladaptive process mediating heightened negative affect in depressed samples. However, this interpretation depends on qualitatively ascribing 'adaptive' labels *versus* 'maladaptive' labels to segregated functioning of the vlPFC *versus* the amygdala respectively, and future research with task data is necessary to corroborate these interpretations.

The present preliminary results may be placed in the context of prior FC analyses in depression. Whereas prior studies have found altered bivariate frontolimbic connectivity (Elliott *et al*. 2011), such as increased connectivity between the amygdala and the OFC (Johnstone *et al*. 2007) and decreased connectivity between the amygdala and the dlPFC (Erk *et al*. 2010), our results focused on network-level FC patterns. Our results replicate prior findings of altered FC of the left amygdala with regions involved in emotion regulation, and we extend prior findings by providing some evidence that susceptibility for depression may be coded by altered network-level FC. Specifically, the preliminary results suggest greater hub-like qualities of the left amygdala within the network and generally decreased connectivity of the left lateral PFC (left dlPFC and left vlPFC) within the network. Further exploration of network-level connectivity may provide a more nuanced understanding of previous bivariate connectivity findings.

Regression analyses provided preliminary evidence that severity of ELS, but not MDD symptoms,

correlated negatively with both degree and betweenness centrality of the left dlPFC. This corresponds with prior research (Heim *et al*. 2000, 2004) suggesting that altered stress reactivity commonly found among depressed individuals is often more strongly linked with ELS than depression. Indeed, the observed group differences of lowered betweenness centrality of this node in the susceptible group seem to be explained by greater ELS severity in the susceptible group. The susceptibility-related decreases in degree and betweenness centrality suggests a diminished role of the dlPFC in directing and biasing information processing throughout the network, which may help to explain why ELS operates as a potent risk factor for MDD.

The present preliminary study is not without limitations. First, we cannot determined whether the altered neural organization patterns represent altered adaptation following ELS or whether they represent pre-morbid differences in network organization prior to ELS occurrence. Second, many group differences in this first study did not survive correction for multiple comparisons, which raises the need for replication among larger independent samples. Third, the sample was restricted to females, which means that the results can probably only be generalized to women and raises the need to test these findings among men and mixed-sex samples. Fourth, the use of a resting-state scan precludes the ability to correlate neural organization patterns with concurrent task performance, which hinders the ability to infer that certain organization patterns are adaptive *versus* maladaptive. Finally, the resilient group was represented by a small sample of well-characterized individuals who were exposed to less ELS than the susceptible group and may therefore represent a lower dose group.

Declaration of Interest

C. B. Nemeroff: *Research/Grants*: NIH; Agency for Healthcare Research and Quality (AHRQ). *Consulting*: Xhale, Takeda. *Stockholder*: CeNeRx BioPharma; NovaDel Pharma Inc.; PharmaNeuroBoost; Revaax Pharma; Xhale. *Other financial interests*: CeNeRx BioPharma; PharmaNeuroBoost. Patents: Method and devices for transdermal delivery of lithium (US 6375990B1); Method of assessing antidepressant drug therapy via transport inhibition of monoamine neurotransmitters by *ex vivo* assay (US 7148027B2). *Scientific Advisory Boards*: American Foundation for

References

Albert R, Jeong H, Barabasi AL (2000). Error and attack tolerance of complex networks. *Nature* 406, 378–382.

Alstott J, Breakspear M, Hagmann P, Cammoun L, Sporns O (2009). Modeling the impact of lesions in the human brain. *PLoS Computational Biology* 5, e1000408.

Anand A, Li Y, Wang Y, Wu J, Gao S, Bukhari L, Mathews VP, Kalnin A, Lowe MJ (2005). Activity and connectivity of brain mood regulating circuit in depression: a functional magnetic resonance study. *Biological Psychiatry* 57, 1079–1088.

Aron AR, Behrens TE, Smith S, Frank MJ, Poldrack RA (2007). Triangulating a cognitive control network using diffusion-weighted magnetic resonance imaging (MRI) and functional MRI. *Journal of Neuroscience* 27, 3743–3752.

Aron AR, Poldrack RA (2006). Cortical and subcortical contributions to Stop signal response inhibition: role of the subthalamic nucleus. *Journal of Neuroscience* 26, 2424–2433.

Aron AR, Robbins TW, Poldrack RA (2004). Inhibition and the right inferior frontal cortex. *Trends in Cognitive Sciences* 8, 170–177.

Bernstein DP, Fink L, Handelsman L, Foote J, Lovejoy M, Wenzel K, Sapareto E, Ruggiero J (1994). Initial reliability and validity of a new retrospective measure of child abuse and neglect. *American Journal of Psychiatry* 151, 1132–1136.

Bremner JD, Narayan M, Anderson ER, Staib LH, Miller HL, Charney DS (2000a). Hippocampal volume reduction in major depression. *American Journal of Psychiatry* 157, 115–118.

Bremner JD, Vermetten E, Mazure CM (2000b). Development and preliminary psychometric properties of an instrument for the measurement of childhood trauma: the Early Trauma Inventory. *Depression and Anxiety* 12, 1–12.

Bullmore E, Sporns O (2009). Complex brain networks: graph theoretical analysis of structural and functional systems. *Nature Reviews Neuroscience* 10, 186–198.

Bullmore ET, Bassett DS (2011). Brain graphs: graphical models of the human brain connectome. *Annual Review of Clinical Psychology* 7, 113–140.

Carpenter LL, Gawuga CE, Tyrka AR, Lee JK, Anderson GM, Price LH (2010). Association between plasma IL-6 response to acute stress and early-life adversity in healthy adults. *Neuropsychopharmacology* 35, 2617–2623.

Craddock RC, Holtzheimer 3rd PE, Hu XP, Mayberg HS (2009). Disease state prediction from resting state functional connectivity. *Magnetic Resonance in Medicine* 62, 1619–1628.

Craig AD (2002). How do you feel? Interoception: the sense of the physiological condition of the body. *Nature Reviews Neuroscience* 3, 655–666.

Critchley HD, Wiens S, Rotshtein P, Ohman A, Dolan RJ (2004). Neural systems supporting interoceptive awareness. *Nature Neuroscience* 7, 189–195.

Davidson RJ, Irwin W, Anderle MJ, Kalin NH (2003). The neural substrates of affective processing in depressed patients treated with venlafaxine. *American Journal of Psychiatry* 160, 64–75.

Davis M, Whalen PJ (2001). The amygdala: vigilance and emotion. *Molecular Psychiatry* 6, 13–34.

Deco G, Jirsa VK, McIntosh AR (2011). Emerging concepts for the dynamical organization of resting-state activity in the brain. *Nature Reviews Neuroscience* 12, 43–56.

Elliott R, Zahn R, Deakin JF, Anderson IM (2011). Affective cognition and its disruption in mood disorders. *Neuropsychopharmacology* 36, 153–182.

Erk S, Mikschl A, Stier S, Ciaramidaro A, Gapp V, Weber B, Walter H (2010). Acute and sustained effects of cognitive emotion regulation in major depression. *Journal of Neuroscience* 30, 15726–15734.

First MB, Spitzer Robert L, Gibbon M, Williams JBW (2002). *Structured Clinical Interview for DSM-IV-TR Axis I Disorders, Research Version, Non-Patient Edition*. Biometrics Research, New York State Psychiatric Institute: New York.

Franco A, Shin J, Choi KS, Craddock R, Mayberg H, Hu X (2011). Increased intra-slice functional correlation: problem and solution. Poster presented at the annual Organization for Human Brain Mapping Conference. Quebec City, Canada.

Gillespie CF, Nemeroff CB (2007). Corticotropin-releasing factor and the psychobiology of early-life stress. *Current Directions in Psychological Science* 16, 85–89.

Greicius MD, Flores BH, Menon V, Glover GH, Solvason HB, Kenna H, Reiss AL, Schatzberg AF (2007). Resting-state functional connectivity in major depression: abnormally increased contributions from subgenual cingulate cortex and thalamus. *Biological Psychiatry* 62, 429–437.

Hamilton M (1960). A rating scale for depression. *Journal of Neurology, Neurosurgery, and Psychiatry* 23, 56–62.

Hanson JL, Chung MK, Avants BB, Shirtcliff EA, Gee JC, Davidson RJ, Pollak SD (2010). Early stress is associated with alterations in the orbitofrontal cortex: a tensor-based morphometry investigation of brain structure and behavioral risk. *Journal of Neuroscience* 30, 7466–7472.

Heberlein KA, Hu X (2004). Simultaneous acquisition of gradient-echo and asymmetric spin-echo for single-shot z-shim: Z-SAGA. *Magnetic Resonance in Medicine* 51, 212–216.

Heim C, Newport DJ, Heit S, Graham YP, Wilcox M, Bonsall R, Miller AH, Nemeroff CB (2000). Pituitary-adrenal and autonomic responses to stress in women after sexual and physical abuse in childhood. *Journal of the American Medical Association* 284, 592–597.

Heim C, Plotsky PM, Nemeroff CB (2004). Importance of studying the contributions of early adverse experience to neurobiological findings in depression. *Neuropsychopharmacology* 29, 641–648.

James GA, Kelley ME, Craddock RC, Holtzheimer PE, Dunlop BW, Nemeroff CB, Mayberg HS, Hu XP (2009). Exploratory structural equation modeling of resting-state fMRI: applicability of group models to individual subjects. *NeuroImage* **45**, 778–787.

Johnstone T, Van Reekum CM, Urry HL, Kalin NH, Davidson RJ (2007). Failure to regulate: counterproductive recruitment of top-down prefrontal-subcortical circuitry in major depression. *Journal of Neuroscience* **27**, 8877–8884.

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

Kilpatrick DG, Ruggiero KJ, Acierno R, Saunders BE, Resnick HS, Best CL (2003). Violence and risk of PTSD, major depression, substance abuse/dependence, and comorbidity: results from the National Survey of Adolescents. *Journal of Consulting and Clinical Psychology* **71**, 692–700.

LeDoux JE (2000). Emotion circuits in the brain. *Annual Review of Neuroscience* **23**, 155–184.

Lee BT, Seok JH, Lee BC, Cho SW, Yoon BJ, Lee KU, Chae JH, Choi IG, Ham BJ (2008). Neural correlates of affective processing in response to sad and angry facial stimuli in patients with major depressive disorder. *Progress in Neuropsychopharmacology and Biological Psychiatry* **32**, 778–785.

Lynall ME, Bassett DS, Kerwin R, McKenna PJ, Kitzbichler M, Muller U, Bullmore E (2010). Functional connectivity and brain networks in schizophrenia. *Progress in Neuropsychopharmacology and Biological Psychiatry* **30**, 9477–9487.

Matthews SC, Strigo IA, Simmons AN, Yang TT, Paulus MP (2008). Decreased functional coupling of the amygdala and supragenual cingulate is related to increased depression in unmedicated individuals with current major depressive disorder. *Journal of Affective Disorders* **111**, 13–20.

Matz K, Junghofer M, Elbert T, Weber K, Wienbruch C, Rockstroh B (2010). Adverse experiences in childhood influence brain responses to emotional stimuli in adult psychiatric patients. *International Journal of Psychophysiology* **75**, 277–286.

McCauley J, Kern DE, Kolodner K, Dill L, Schroeder AF, Dechant HK, Ryden J, Derogatis LR, Bass EB (1997). Clinical characteristics of women with a history of childhood abuse: unhealed wounds. *Journal of the American Medical Association* **277**, 1362–1368.

McRae K, Hughes B, Chopra S, Gabrieli JD, Gross JJ, Ochsner KN (2010). The neural bases of distraction and reappraisal. *Journal of Cognitive Neuroscience* **22**, 248–262.

Mueller SC, Maheu FS, Dozier M, Peloso E, Mandell D, Leibenluft E, Pine DS, Ernst M (2010). Early-life stress is associated with impairment in cognitive control in adolescence: an fMRI study. *Neuropsychologia* **48**, 3037–3044.

Ochsner KN, Bunge SA, Gross JJ, Gabrieli JD (2002). Rethinking feelings: an fMRI study of the cognitive regulation of emotion. *Journal of Cognitive Neuroscience* **14**, 1215–1229.

Ochsner KN, Ray RD, Cooper JC, Robertson ER, Chopra S, Gabrieli JD, Gross JJ (2004). For better or for worse: neural systems supporting the cognitive down- and up-regulation of negative emotion. *NeuroImage* **23**, 483–499.

Price JL, Drevets WC (2010). Neurocircuitry of mood disorders. *Neuropsychopharmacology* **35**, 192–216.

Resnick HS, Kilpatrick DG, Dansky BS, Saunders BE, Best CL (1993). Prevalence of civilian trauma and posttraumatic stress disorder in a representative national sample of women. *Journal of Consulting and Clinical Psychology* **61**, 984–991.

Rubinov M, Sporns O (2010). Complex network measures of brain connectivity: uses and interpretations. *NeuroImage* **52**, 1059–1069.

Seminowicz DA, Mayberg HS, McIntosh AR, Goldapple K, Kennedy S, Segal Z, Rafi-Tari S (2004). Limbic-frontal circuitry in major depression: a path modeling metanalysis. *NeuroImage* **22**, 409–418.

Taylor SE, Eisenberger NI, Saxbe D, Lehman BJ, Lieberman MD (2006). Neural responses to emotional stimuli are associated with childhood family stress. *Biological Psychiatry* **60**, 296–301.

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 Wijk BC, Stam CJ, Daffertshofer A (2010). Comparing brain networks of different size and connectivity density using graph theory. *PLoS ONE* **5**, e13701.

Vasic N, Walter H, Sambataro F, Wolf RC (2009). Aberrant functional connectivity of dorsolateral prefrontal and cingulate networks in patients with major depression during working memory processing. *Psychological Medicine* **39**, 977–987.

Victor TA, Furey ML, Fromm SJ, Ohman A, Drevets WC (2010). Relationship between amygdala responses to masked faces and mood state and treatment in major depressive disorder. *Archives of General Psychiatry* **67**, 1128–1138.

Vythilingam M, Heim C, Newport J, Miller AH, Anderson E, Bronen R, Brummer M, Staib L, Vermetten E, Charney DS, Nemeroff CB, Bremner JD (2002). Childhood trauma associated with smaller hippocampal volume in women with major depression. *American Journal of Psychiatry* **159**, 2072–2080.

Wager TD, Davidson ML, Hughes BL, Lindquist MA, Ochsner KN (2008). Prefrontal-subcortical pathways mediating successful emotion regulation. *Neuron* **59**, 1037–1050.

Weber K, Miller GA, Schupp HT, Borgelt J, Awiszus B, Popov T, Elbert T, Rockstroh B (2009). Early life stress and psychiatric disorder modulate cortical responses to affective stimuli. *Psychophysiology* **46**, 1234–1243.

Williams LM, Gatt JM, Schofield PR, Olivieri G, Peduto A, Gordon E (2009). 'Negativity bias' in risk for depression and anxiety: brain-body fear circuitry correlates, 5-HTT-LPR and early life stress. *NeuroImage* **47**, 804–814.