# Childhood emotional maltreatment : impact on cognition and the brain

Van Harmelen, A.

# Chapter 4: Reduced medial PreFrontal Cortex volume in adults reporting Childhood Emotional Maltreatment

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**ABSTRACT**

Childhood emotional maltreatment (CEM) has been associated with a profound and enduring negative impact on behavioral and emotional functioning. Animal models have shown that adverse rearing conditions, such as maternal separation, can induce a cascade of long-term structural alterations in the brain, particularly in the hippocampus, amygdala, and prefrontal cortex (PFC). However, in humans, the neurobiological correlates of CEM are unknown. Using high-resolution T1-weighted 3T MRI anatomical scans and a whole-brain optimized Voxel Based Morphometry approach, we examined whether healthy controls and unmedicated patients with depression and/or anxiety disorders reporting CEM before the age of 16 (n=84, age: M=38.7) displayed structural brain changes compared to controls and patients who report no childhood abuse (n=97, age: M=36.6). We found that self-reported CEM is associated with a significant reduction in predominantly left dorsal medial prefrontal cortex (dmPFC) volume, even in the absence of physical and/or sexual abuse during childhood. In addition, reduced dmPFC in individuals reporting CEM is present in males and females, and independent of concomitant psychopathology. In this study, we show that CEM is associated with profound reductions of mPFC volume, suggesting that sustained inhibition of growth or structural damage can occur after exposure to CEM. Given the important role of the mPFC in the regulation of emotional behavior, our finding might provide an important link in understanding the increased emotional sensitivity in individuals reporting CEM.

## INTRODUCTION

Every year, about one in ten children growing up in Western societies experiences Childhood Emotional Maltreatment (CEM; Egeland, 2009; Gilbert et al., 2009). Emotional maltreatment encompasses any act of commission (i.e. verbal abuse) or omission (i.e. emotional neglect) that is (potentially) harmful, or insensitive to the child's emotional development (Egeland, 2009; Gilbert et al., 2009), and has been associated with a profound and enduring negative impact on behavioral, emotional, and social functioning (Egeland, 2009; Gilbert et al., 2009). For instance, CEM is associated with maladaptive emotional functioning in adulthood (Teicher, Samson, Polcari, & McGreenery, 2006), which in turn is a key vulnerability factor for the development of psychiatric disorders when faced with stressors in later life (Beck, 2008). In line with this, CEM is an important predictor for the development of depressive and anxiety disorders in adulthood (Gibb, Benas, Crossett, & Uhrlass, 2007; Spinhoven et al., 2010). However, the neurobiological correlates underlying the emotional sensitivity in individuals reporting CEM are yet unknown.

In animals, adverse rearing environments such as maternal separation, loss, or isolation rearing induce a cascade of long-term alterations on the level of cognitive functioning, hypothalamic-pituitary (HPA) axis functioning, (immediate) gene expression and brain morphology (Sánchez, Ladd, & Plotsky, 2001). Structural alterations in the brain include reduced dendrite length, dendritic branching, spine density, and suppression of neurogenesis, and have predominantly been observed in limbic structures (amygdala, hippocampus) and prefrontal cortex (PFC)(Arnsten, 2009; Lupien, McEwen, Gunnar, & Heim, 2009; McEwen, 2008; Sánchez et al., 2001). In line, human studies examining the neuroanatomical correlates of childhood maltreatment in adults found decreased gray matter (GM) volume in the hippocampus (Kitayama, Vaccarino, Kutner, Weiss, & Bremner, 2005; Vythilingam et al., 2002), and medial (m)PFC (Andersen et al., 2008; Cohen et al., 2006; Frodl et al., 2010; Tomoda et al., 2009; Treadway et al., 2009). However, these studies focused mainly on the impact of sexual (Andersen et al., 2008; Kitayama et al., 2005; Vythilingam et al., 2002) and/or physical abuse (Tomoda et al., 2009), or did not exclude co-occurrence of different types of abuse (Cohen et al., 2006; Frodl et al., 2010; Treadway et al., 2009).

One way through which chronic stress may lead to structural changes is by means of enhanced activation of neuroendocrine systems (McEwen, 2008). During chronic stress, increased secretion of glucocorticoids (i.e. the stress hormone cortisol in humans) interferes with the transcriptional mechanisms that control the expression of brain-derived neurotrophic factor (BDNF), a growth factor that has been linked to neuronal proliferation and plasticity (McEwen, 2008; Nestler et al., 2002). In this way, chronic stress may inhibit cytogenesis and increase vulnerability to attrition within the hippocampus, amygdala and/or PFC (e.g. Lupien et al., 2009; McEwen, 2008). In line with these findings, childhood maltreatment has been linked to enhanced cortisol reactivity to psychosocial stress in patients with

depression and anxiety disorders (Elzinga, Spinhoven, Berretty, De Jong, & Roelofs, 2010; Heim, 2000; Heim et al., 2002) and to blunted cortisol reactivity in healthy subjects (Carpenter et al., 2007). Additionally, altered patterns of cortisol reactivity during stress have been found in individuals reporting CEM (Carpenter et al., 2009; Elzinga et al., 2010). Furthermore, white matter (WM) tract abnormalities were found in a small sample of young adults reporting CEM (n =16; Choi, Jeong, Rohan, Polcari, & Teicher, 2009). However, it is yet unknown whether CEM is similarly associated with structural GM abnormalities in adulthood. Given the important role of the limbic brain regions (hippocampus and amygdala) and the mPFC in the perception and regulation of emotional behavior and stress response (Arnsten, 2009; Cardinal, Parkinson, Hall, & Everitt, 2002; Lupien et al., 2009; McEwen, 2008), GM abnormalities in (one of) these regions might underlie the maladaptive emotional functioning associated with CEM.

Therefore, we sought to investigate the effect of CEM on adult brain morphology in unmedicated patients currently diagnosed with depression and/or anxiety disorder and healthy controls (HCs). We used high resolution Magnetic Resonance Imaging (MRI) and a whole-brain optimized Voxel Based Morphometry analysis approach, and specified the amygdala, hippocampus, and mPFC (medial prefrontal gyrus and anterior cingulate gyrus) as Regions of interest (ROI). We examined whether adult patients and HCs reporting multiple incidents of CEM before the age of 16 (n=84) displayed structural brain changes in comparison to patients and HCs who did not report a history of childhood abuse (n=97). In addition, to examine whether these structural brain changes are related to the development of psychopathology, we investigated whether these brain alterations were more apparent in individuals with a depression and/or anxiety disorder compared to individuals who never developed a depression and/or anxiety disorder.

## METHODS

### THE NESDA –MRI STUDY

Participants were drawn from the Netherlands Study of Depression and Anxiety (NESDA), (N=2981), a large cohort study (Penninx et al., 2008). A subset of the NESDA participants (both patients and HCs) was selected to undergo MRI scanning for the NESDA MRI study. Inclusion criteria for patients in the NESDA-MRI study were: current major depressive disorder (MDD) and/or anxiety disorder (ANX; panic disorder (PD) and/or social anxiety disorder (SAD) and/or Generalized Anxiety Disorder (GAD) in the last 6 months according to DSM-IV criteria). Diagnoses were established using the structured Composite International Diagnostic Interview (CIDI: Wittchen et al., 1991), administered by a trained interviewer. Exclusion criteria were: the presence of axis-I disorders other than MDD, PD, SAD or GAD; any use of psychotropic medication other than a stable use of selective serotonin reuptake inhibitors (SSRI) or infrequent benzodiazepine use [3

times 2 tablets weekly, or within 48 hrs prior to scanning]. Additional exclusion criteria for both patients and HCs were: the presence or history of major internal or neurological disorder; dependency or recent abuse [past year] of alcohol and/or drugs; hypertension (>180/130mm Hg); heavy smoker (>5 cigarettes/day); and general MR-contra indications. HCs had no lifetime depressive or anxiety disorders and were not taking any psychotropic drugs. Eventually, 301 native Dutch speaking participants (235 patients and 66 HCs) were included and underwent MR imaging in one of the three participating centers (i.e. Leiden University Medical Center [LUMC], Amsterdam Medical Center [AMC], and University Medical Center Groningen [UMCG]). The Ethical Review Boards of each participating center approved this study. All participants provided written informed consent.

### Clinical assessments

In the NESDA study, childhood maltreatment was assessed with the Nemesis trauma interview (De Graaf, Bijl, Smit, Vollebergh, & Spijker, 2002). In this interview, respondents were asked whether they had experienced emotional neglect, psychological abuse, physical abuse and/or sexual abuse before the age of 16, how often this had occurred (i.e. never, once, sometimes, regularly, often, or very often), and what their relationship with the perpetrator was. Emotional neglect was described as: 'people at home didn't listen to you, your problems were ignored, and you felt unable to find any attention or support from the people in your house'. Psychological abuse was described as: 'you were cursed at, unjustly punished, your brothers and sisters were favoured – but no bodily harm was done'. CEM was defined as multiple incidents (>once) of emotional neglect and/or emotional abuse before the age of 16 years, because we assumed that only multiple incidents of emotional abuse and/or emotional neglect might be associated with neuroanatomical changes. Overall CEM frequency was defined as the most frequent occurrence as reported (e.g. if psychological abuse occurred often, and emotional neglect sometimes, overall CEM score is often).

Negative life events were assessed using the List of Threatening Events Questionnaire (LTE-Q; Brugha, Bebbington, Tennant, & Hurry, 1985). In addition, at the day of scanning, depression and anxiety severity was measured using the Montgomery Åsberg Depression Rating Scale (MADRS; Montgomery & Asberg, 1979) and the Becks Anxiety Inventory (BAI; Beck, Epstein, Brown, & Steer, 1988).

### Additional exclusion criteria

High resolution anatomical images were obtained from 291 participants (imaging data from 10 participants were excluded because of poor image quality). Additionally, two healthy controls were excluded from the NESDA-MRI study because of MADRS scores that were indicative of possible depressive symptomatology at the day of scanning (MADRS>8; Müller, Szegedi, Wetzel, & Benkert, 2000). For the present study, individuals using SSRIs were excluded (n=79) given their potential effect on neuronal

plasticity (e.g. Dranovsky & Hen, 2006). Additionally, individuals reporting physical or sexual abuse but no CEM were also excluded (n=5). Finally, individuals reporting only a single incident of CEM (n=24) were excluded. Our final sample (n=181) consisted of 84 participants reporting CEM, and 97 participants that reported No Abuse.

### THE CEM AND NO ABUSE GROUPS

The CEM group consisted of participants who reported emotional neglect or psychological abuse during childhood hat had occurred sometimes, regularly, often or very often (n=84, i.e. MDD (n=20), ANX (n=21), Comorbid Depression and Anxiety disorder ([CDA] n=30), and HC (n=13), of whom 36 participants also reported childhood physical and/ or sexual abuse, see Table 1). The No Abuse group consisted of individuals who did not report CEM, physical or sexual abuse (n=97; i.e. MDD (n=22), ANX (n=22), CDA (n=13), and HC (n=40)). In the CEM group, 96.4 % (n=81) of the participants reported emotionally neglect, whereas 57.1 % (n=48) reported to have been psychologically abused, and 54% (n=45) reported both emotional neglect and psychological abuse. In addition, 97.6 % reported that the individual's biological parents were the perpetrators of CEM (n=82).

**Table 1.** Clinical and demographic characteristics of participants reporting Childhood Emotional Maltreatment vs. No Abuse

| | | No Abuse (*n*= 97) | CEM (*n*= 84) | F | U | $X^2$ | P |
|---|---|---|---|---|---|---|---|
| **Gender** | % M/ F | 33/ 67 | 34.5/ 65.5 | | | .05 | .83 |
| **Age** | Mean (SEM) | 36.57 (1.09) | 38.68 (1.09) | 1.86 | | | .17 |
| **Education** | Mean (SEM) | 13.27 (0.29) | 12.81 (0.35) | | 3706.5 | | .29 |
| **Handedness** | % L/ R | 11/ 89 | 5/95 | | | 2.56 | .11 |
| **Current diagnosis** | *n* MDD | 22 | 20 | | | .09 | .76 |
| | *n* ANX | 22 | 21 | | | .02 | .88 |
| | *n* CDA | 13 | 30 | | | 6.72 | .01 |
| | *n* HC | 40 | 13 | | | 13.75 | .00 |
| **Lifetime diagnosis** | % MDD | 43.29 | 77.38 | | | 25.64 | .00 |
| | % ANX | 41.24 | 67.86 | | | 12.83 | .00 |
| **MADRS** | Mean (SEM) | 7.10 (.94) | 14.45 (1.89) | | 2272.5 | | .00 |
| **BAI** | Mean (SEM) | 8.79 (1.04) | 12.85 (1.08) | | 2651.5 | | .00 |
| **Scan location** | % A/ L/ G | 28.9/ 41.2/ 29.9 | 38.1/ 38.1/ 23.8 | | | 1.89 | .39 |
| **Frequency of CEM** | %S/R/O/V | 0 | 10.8/ 37.4/ 21.7/ 30.1 | | | | |
| **Concurrent abuse** | *n* Physical | 0 | 15 | | | | |
| | *n* Sexual abuse | 0 | 8 | | | | |
| | *n* Physical and Sexual abus | 0 | 13 | | | | |
| **Gray Matter** | Mean (SEM) | 740.40 (7.98) | 721.78 (7.33) | 2.89 | | | .09 |
| **White matter** | Mean (SEM) | 491.33 (6.94) | 494.69 (6.53) | .12 | | | .73 |

*Note*. CEM= Childhood Emotional Maltreatment, ANX= Anxiety Disorder, MDD= Major Depressive Disorder, CDA = Comorbid Depression and Anxiety Disorder, S=sometimes, R=regularly, O=often, V= very Often, A= Amsterdam Medical Center, L= Leiden University Medical Center, G= University Medical Center Groningen, MADRS= Montgomery Åsberg Depression Rating Scale, BAI= Beck Anxiety Inventory, F,U, $X^2$ = F ratio, Mann Whitney U statistic, and Chi-square test statistic, SEM= Standard Error of Mean.

## MRI

### Image Acquisition

Imaging data were acquired using Philips 3T MR-systems located at the participating centers, equipped with a SENSE-8 (LUMC and UMCG) and a SENSE-6 (AMC) channel head coil. For each subject an anatomical image was obtained using a sagittal 3D gradient-echo T1-weighted sequence (TR: 9 ms; TE: 3.5 msec; matrix 256×256; voxel size: 1×1×1 mm; 170 slices). Each scanning session also included several fMRI runs, both 'resting-state' and task-related. These results, as well as those of VBM comparisons between diagnostic groups (irrespective of childhood maltreatment), will be reported elsewhere.

### Image preprocessing

An optimized Voxel Based Morphometry (VBM) approach following the Diffeomorphic Anatomical Registration Through Exponentiated Lie algebra (DARTEL; Ashburner, 2007) was performed using SPM5 (Statistical Parametric Mapping software; http:// www.fil.ion.ucl.ac.uk/spm/) implemented in Matlab 7.1.0 (The MathWorks Inc., MA, USA). VBM-DARTEL preprocessing included the following steps; 1) manually reorientation of the images to the anterior commissure, 2) segmentation of the anatomical images using the standard segmentation option implemented in SPM5, 3) applying the DARTEL approach for registration, normalization, and modulation, leaving the images in DARTEL space. In this approach, a DARTEL template is created based on the deformation fields that are produced by the segmentation procedure. Next, all individual deformation fields are warped (and modulated) to match this template. 4) Smoothing of the gray matter (GM) and white matter (WM) images using an 8mm, full width at half maximum, Gaussian kernel to increase signal to noise ratio. In the resulting GM images, each voxel represents an absolute amount of GM volume, equivalent to the GM volume per unit prior to normalization.

### VBM analysis

GM (or WM) segments in native space were used to calculate absolute total GM (or WM) volumes. Next, smoothed GM (WM) density images were entered into a voxel by voxel analysis of variance for between-group comparisons, with age and total absolute GM (or WM) as covariate to correct for total brain volume. Effect of center was added by means of two dummy variables as extra regressors in all analyses. To get maximal sensitivity, and to optimize voxel residual smoothness estimation and to exclude false positives in non-GM (or WM) tissue, voxel-wise comparisons were masked using a comparison-specific explicit optimal threshold GM (or WM) mask created using the Masking toolbox (Ridgway et al., 2009).

For the a priori set ROIs (mPFC, amygdala and hippocampus), we set a threshold of $P<.001$, uncorrected, with a spatial extent threshold of 50 contiguous voxels for group interactions. To further protect against Type I error, Small Volume Correction (SVC) was applied, by centering a sphere of

16 mm around the peak voxel. The resulting Volumes of Interest had to meet P<.05, FWE voxel corrected, to be considered significant (i.e. Z>3.50). For regions not a priori specified, a voxel level threshold of P<.05 whole brain FWE corrected was set. If significant group differences were observed in the VBM analysis, we then exported the volume of the significant clusters (i.e. K centered around the peak voxel) per subject to SPSS. Clinical and demographic group differences were analyzed using SPSS-17 (www.spss.com), and in all analyses, age, total GM (WM) volume, and dummy regressors for the scan centers were included as covariates.

## RESULTS

### THE NEUROANATOMICAL CORRELATES OF CHILDHOOD EMOTIONAL MALTREATMENT

To investigate the neuroanatomical correlates of CEM, we first set up a VBM analysis to compare the GM density maps/images of individuals reporting CEM (n=84) with GM density maps of the No Abuse group (n=97). These analyses revealed that CEM was associated with a 5.14 % reduction in the left dorsal mPFC (x=-11 y=23 z=40, Brodmann Area (BA) 8, cluster size/ number of voxels (K) =263, Z=3.80, P<.05, Small Volume Corrected (SVC), Table 2). No significant differences were observed in hippocampus or amygdala, or in other brain regions. Only at a very low threshold was CEM was associated with reduced right posterior hippocampal volume (x=29 y=-35 z=-6, Z=2.06, n.s). Additionally, CEM was not associated with a significant increase in regional GM volumes. Finally, CEM was not associated with WM reductions in or surrounding our ROIs, or with increased regional GM volume.

To explore possible interactions between CEM, current diagnosis, and gender, a 2 (CEM)×4 (diagnosis: MDD, ANX, CDA, and HC)×2 (gender) univariate ANCOVA was performed, with local mPFC volume (ml.) as a dependent factor. Again, individuals from the CEM group had smaller mPFC volumes than the No Abuse group (CEM (M± SEM): 4.80 ± .06 ml. vs. No Abuse; 5.06 ± .06 ml. (F(1, 161)= 12.36, P<.01, Cohen's d (d) =.53). Interestingly, there was no interaction between CEM and diagnosis (F(3,161)=. 45, P=.72, d=.01), and post-hoc analyses indicated that the mPFC reductions are present in all groups, even though the numbers were relatively small for such comparisons [one sided: MDD (F(1,34)=8.65, P<.01, d=.93), ANX (F(1,35)=2.55, P=.06, d=.50), CDA (F(1,35)=2.63, P=.06, d=.55), and HC (F(1,45)=1.85, P=.09, d=.44)]. In addition, there was no interaction between CEM and gender (F(1,161)=2.01, P=.99, d=.21). Taken together, these results indicate that reduced mPFC volume was present in all CEM groups (i.e., male and female individuals with MDD, ANX, CDA, and in the HC group). Moreover, similar results were obtained when depression and

anxiety severity were added as covariates (F(1,155)=12.41, P<.01, d=.53)[III], indicating that our results cannot be explained by the presence of more severe depressive and/or anxious symptomatology amongst individuals reporting CEM.

### NEUROANATOMICAL CORRELATES OF ISOLATED EMOTIONAL MALTREATMENT IN CHILDHOOD.

To exclude the possibility that our results are driven by concurrent history of physical and/or sexual abuse in some of the participants, we conducted a whole brain VBM analysis to compare the GM density maps of individuals reporting only CEM (n=48; MDD (n=13), ANX (n=12), CDA (n=13), and HC (n =10) see Table S1) with individuals reporting No Abuse (n=97). In this analysis the 36 individuals who also reported childhood physical and/or sexual abuse were excluded. This analysis showed that individuals reporting only CEM had a volume reduction of 7.2 % in left and right (although predominantly left) dorsal medial mPFC (x=-11 y=21 z=40, BA 8, K=767, Z=4.37, P<.05 (SVC), see Table 2), extending into the anterior mPFC and anterior cingulate gyrus (Figure 1, Table 2).

**Table 2**. The neuroanatomical correlates of the Childhood Emotional Maltreatment vs. No Abuse.

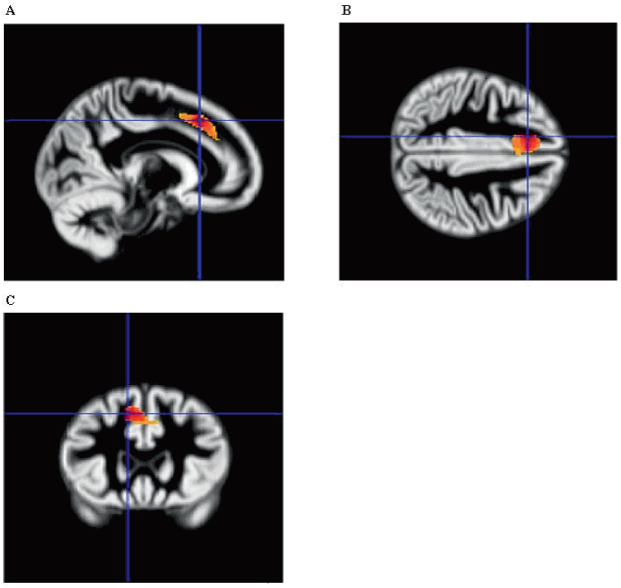
| | | | | | | DARTEL -coordinates | | | |
|---|---|---|---|---|---|---|---|---|---|
| | | R/L | BA | region | K | x | y | z | Z |
| **CEM (*n* =97) vs. No Abuse (*n* =84)** | CEM< No Abuse | L | 8 | Medial prefrontal gyrus | 263 | -11 | 23 | 40 | 3.80 ** |
| **only CEM (n=48) vs. No Abuse (n=97)** | CEM< No Abuse | L+R | 8 | Medial prefrontal gyrus | 767 | -11 | 21 | 40 | 4.37 ** |
| | | | | | | -5 | 9 | 42 | |
| | | | | | | 6 | 18 | 36 | |
| | | R | 9/32 | Cingulate gyrus/medial prefrontal | 87 | 9 | 44 | 16 | 3.53 ** |
| | | R | 10/32 | Cingulate gyrus/medial prefrontal | | 11 | 47 | 9 | |

*Note.* CEM= Childhood Emotional Maltreatment, R=right sided, L=Left sided, BA= Brodmann Area, K= cluster size (number of voxels), **=P<.05, SVC 16 mm FWE corrected.

Furthermore, no hippocampal, or amygdalar differences were observed, nor decreases in other brain regions. Again, only at a very low threshold was CEM associated with reduced right posterior hippocampal volume (x=29 y=-35 z=-6, Z=2.45, n.s). Finally, CEM was not associated with WM reductions in or surrounding our ROIs, or with increased regional GM volume.

[III] Four participants were missing because of incomplete depression or anxiety data (one reported CEM).

**Figure 1**. The medial PFC region showing 7.2 % volume reduction amongst individuals reporting only childhood emotional maltreatment displayed on a sagittal (A), transversal (B) and coronal (C) plane.



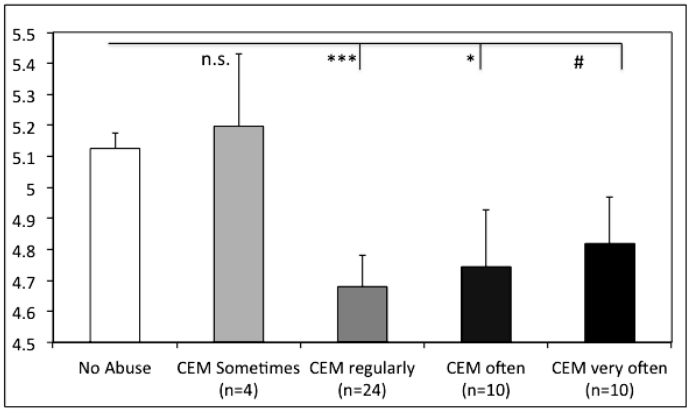

An ANCOVA confirmed the main effect of CEM (i.e. CEM: M ± SEM: 4.78 ± .07 ml., No Abuse: 5.15 ± .06 ml., (F(1,125=15.15, P<.001, d=.69). Moreover, the reduced mPFC volume was present in all CEM groups and in both male and female individuals (i.e. no interaction was found with diagnosis (F(3,125)=.27, P=.85, d=.09), and even within these small groups post-hoc analyses revealed a (marginally) significant (one–sided) impact of CEM only on mPFC volume ((i.e. MDD, (F(1,27)=7.72, P<.01, d=1), ANX (F(1,26)=2.93, P<.05. d=.63), CDA (F(1,18)=3.20, P<.05, d=.73), and HC (F(1,42)=1.96, P=.08, d=.51)). Moreover, CEM did not interact with gender (F(1,125)=.08, P=.78, d=.05). These results could not be explained by higher symptom severity amongst individuals reporting CEM, since similar results were obtained when depression and anxiety severity were added as covariates (F(1,116)=15.72, P<.001, d=.70[IV]).

[IV] *History of alcoholism (abuse or dependence as measured with the CIDI) did not affect the results (i.e. history of alcoholism (yes or no) was not a significant covariate in the analysis (F(1,124) =1.76, P=.19), nor did it impact the main effect of CEM on mPFC volume (F(1,124) =15.98, P<.000, d=.71)).*

### ASSOCIATIONS BETWEEN FREQUENCY OF EMOTIONAL MALTREATMENT AND MPFC VOLUME

To investigate whether the mPFC volume reductions were dependent on CEM frequency, we performed a 5 (frequency of CEM: no abuse, sometimes, regularly, often, and very often) × 4 (diagnosis: MDD, ANX, CDA, and HC) ANCOVA, with local mPFC volume (ml.) as a dependent factor. The analysis revealed a main effect of frequency of CEM on mPFC volume ($F(4,120)=4.89$, $P<.001$, $d=.39$)-, which did not interact with psychopathology ($F(12,120)=.93$, $P=.52$, $d=.17$). As is illustrated in Figure 2, mPFC volumes were reduced in individuals reported that CEM happened regularly or more often. Individuals reporting CEM sometimes did not have a significantly lower mPFC volume when compared to the No Abuse group, however, this group was extremely small (n=4)[V] therefore caution is warranted when interpreting the findings of these individuals.

**Figure 2.** Estimated Marginal Means and Standard Error of Mean (SEM) of medial PreFrontal Cortex (mPFC) volume amongst the different frequencies of Childhood Emotional Maltreatment (CEM), and contrast results of CEM frequencies vs. the No Abuse group.




***Note.*** ***= P<.000, * = P<.05, # = P=.056, n.s. = not significant (two-sided).

[V] *Exclusion of the 'sometimes' group (n=4), due to its small size, did not affect the results, including the main effect of CEM on mPFC volume ($F(1,120)=15.8$, $P<.000$, $d=.73$).*

## DISCUSSION

In this study, self-reported CEM was found to be associated with a significant reduction in predominantly left dorsal mPFC GM volume, independent of gender, and psychiatric status, at least in individuals who reported CEM regularly, or more frequent. Furthermore, the mPFC GM volume reduction was not due to concomitant childhood physical and/or sexual abuse, as the reductions were also found when CEM was experienced in absence of concurrent childhood physical and/or sexual abuse.

These findings provide an important clinical extension of pre-clinical observations that the mPFC is highly sensitive to the effects of chronic stress in childhood (Arnsten, 2009; Lupien et al., 2009; McEwen, 2008). The mPFC is one of the brain regions that undergo major developmental changes during childhood and adolescence (Arnsten, 2009; Lupien et al., 2009). Exposure to emotionally abusive episodes during this developmental time period may increase secretion of glucocorticoids, which may interfere with the transcriptional mechanisms that control the expression of BDNF, and may thereby inhibit cytogenesis and increase vulnerability to attrition within the mPFC (Arnsten, 2009; Lupien et al., 2009; McEwen, 2008; Nestler et al., 2002). Moreover, the fact that reductions in hippocampal volume were only observed at a very low threshold, and no significant changes were observed in the amygdala, concurs with findings of animal models on isolation rearing or maternal separation that indicate a specific and profound impact on the mPFC (Levine et al., 2008; Sanchez et al., 2007), in comparison to the hippocampus and amygdala (Schubert, Porkess, Dashdorj, Fone, & Auer, 2009). For example, in animals, it has been shown that architectural changes in prefrontal dendrites can already be observed after only one week of stress, or even after a single stressful incident (Arnsten, 2009). In contrast, structural changes in the hippocampus only appear after several weeks of stress, which might be an indication that the mPFC is more sensitive to the detrimental effects of stress (Arnsten, 2009).

The finding that CEM is associated with (predominantly left) dorsal mPFC reduction is of particular interest when considering the fact that the mPFC plays an important role in emotion regulation (Cardinal et al., 2002; Phillips, Drevets, Rauch, & Lane, 2003). Moreover, reduced activity in the left PFC has been particularly associated with negative emotional states (Demaree, Everhart, Youngstrom, & Harrison, 2005). Furthermore, the dorsal mPFC is essential for the regulation of autonomic and neuroendocrine stress response and arousal associated with emotional states and behavior, while the ventral mPFC has been implicated in generating these emotional states and behaviour (Phillips et al., 2003; Radley, Williams, & Sawchenko, 2008). The dorsal and ventral mPFC are reciprocally functionally related, and abnormalities in the function of either, or both, may be associated with abnormalities in emotional behavior and regulation (Phillips et al., 2003). In line with these findings, decreased blood flow in the dorsal mPFC has been associated with increased autonomic responsiveness, anxiety, and sad mood

(Phillips et al., 2003). In addition, mPFC dysfunctions have been implicated in many psychiatric disorders, including depressive disorders (Drevets, Price, & Furey, 2008) and anxiety disorders (Zhao et al., 2007). Taken together, these results suggest that the reduced dorsal mPFC volume may (partly) underlie the enhanced emotional sensitivity associated with CEM. It should be noted that, contrary to our predictions, the reduced mPFC volume associated with CEM was independent of psychopathological status, indicating that the reduced mPFC volume was not only present in individuals with psychopathology, but also in HCs who never developed a depression or anxiety disorder (even though the number of HCs with reported CEM is relatively small, and effect sizes of mPFC reductions were also smaller in the HCs than in individuals with depression and/or anxiety). Therefore, reduced mPFC volume does not seem to be directly linked to the development of depressive and/or anxiety disorders in individuals reporting CEM. This finding is more consistent with the idea that additional risk factors, such as genetic make-up (Frodl et al., 2010; Gatt et al., 2009; Joffe et al., 2009) alone, or in interaction with exposure to stressful life events during adulthood may additionally determine who will subsequently develop a depressive and/or anxiety disorder (Beck, 2008; Caspi & Moffitt, 2006). In line with this suggestion, individuals with current depressive and/or anxiety disorder reporting CEM (n=65) indeed reported more negative life events (Mean ± SEM: 5.96 ±.55) than HCs reporting CEM (n=13; 4.62 ±.24), ($t(76)=-2.26$, $P<.05$).

Although the present results are compelling, several potential limitations must be taken into account. The use of (DARTEL-) VBM approaches is not without its limitations (Ridgway et al., 2008), although recent studies (McLaren, Kosmatka, Kastman, Bendlin, & Johnson, 2010; Yassa & Stark, 2009) demonstrated that the DARTEL approach is an improvement to standard voxel-based approaches. In addition, the sensitivity of the DARTEL approach for detecting hippocampal atrophy has been demonstrated in MDD patients (Bergouignan et al., 2009). Nevertheless, manual tracing or shape based analyses techniques, as have been used in most previous studies on hippocampal structural abnormalities, might be more sensitive in detecting deformations compared to an automated segmentation approach. Furthermore, although a clinically diagnosed PTSD diagnosis was an exclusion criterion for NESDA, unidentified current or lifetime PTSD symptoms may still have been present, which may have influenced our findings. However, CEM related mPFC GM reductions were also present amongst HCs who had never developed a depressive or anxiety disorder; therefore, it is unlikely that current or lifetime PTSD may have confounded our results. In addition, history of childhood maltreatment was retrospectively assessed by means of an interview, and it is important to acknowledge the inherent subjectivity of self-reported CEM. For instance, the retrospective assessment of CEM may be subject to recollection bias, so that individuals with current psychopathology may over-report, whereas HCs may under-report a history of childhood maltreatment. However, we

would like to note that in the NESDA sample, current affective state did not moderate the association between CEM and lifetime affective disorder, indicating that recall of CEM in the current sample was not critically affected by current mood state (Spinhoven et al., 2010). Finally, our findings are based on a cross-sectional study. Whereas the idea that CEM is associated with mPFC GM volume reductions fits very well with numerous preclinical studies, the possibility of reversed causality cannot be excluded. For instance, individuals with reduced mPFC volume might report more CEM as a result of impaired emotion regulation. Another explanation may be that the reduced mPFC volume was pre-existent, and that inadequate emotion regulation associated with reduced mPFC volume might even increase children's risk for exposure to CEM. Following this line of thought, one would expect that reports of presence or absence of life stressors later in life would also be related to mPFC volume. Nevertheless, presence of life stressors (yes/no) was not associated with mPFC volume ($F(1,109)=.09$, $P=.76$, $d=.05$), and the impact of CEM on mPFC volume remained unchanged when adding presence of life events into the analysis ($F(1,109)=9.08$, $P<.01$, $d=.54$). Theoretically, longitudinal studies examining neuroanatomical developmental changes over time amongst individuals reporting CEM are needed to shed more light on the etiology of our findings. To the best of our knowledge, such studies have not yet been conducted, and from an ethical point of view it would be highly problematic to prospectively follow children that are known to be exposed to CEM without interfering in their situation.

## CONCLUSION

We found in a large sample of un-medicated adults that self-reported CEM is associated with a substantial reduction in mPFC GM volume. In line with an accumulating number of animal studies (Levine et al., 2008; Lupien et al., 2009; Sánchez et al., 2001; Sanchez et al., 2007), our finding suggests that sustained inhibition of growth, or even structural damage, can occur after exposure to emotional maltreatment in childhood. In addition, previous studies have shown that CEM is associated with altered HPA axis reactivity to stress (Carpenter et al., 2009; Elzinga et al., 2008), and that CEM is an important predictor for the development of depressive and anxiety disorders in adulthood (Gibb et al., 2007; Spinhoven et al., 2010). Given the important role of the mPFC in the perception and regulation of emotional behavior and stress responses (Arnsten, 2009; Cardinal et al., 2002; Lupien et al., 2009; McEwen, 2008; Phillips et al., 2003; Radley et al., 2008; Sánchez et al., 2001), our finding might provide an important link in understanding the increased emotional sensitivity in individuals reporting CEM.

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**Table S1.** Clinical and demographic characteristics of participants reporting only Childhood Emotional Maltreatment vs. No Abuse.

| | | No Abuse (n=97) | only CEM (n=48) | F | U | $X^2$ | P |
|---|---|---|---|---|---|---|---|
| **Gender** | % M/ F | 33/ 67 | 42/ 58 | | | 1.05 | .30 |
| **Age** | Mean (SEM) | 36.57 (1.09) | 37.6 (1.60) | .29 | | | .59 |
| **Education** | Mean (SEM) | 13.27 (0.29) | 13.56 (0.41) | | 2229 | | .67 |
| **Handedness** | % L/ R | 11/ 89 | 6/ 94 | | | .95 | .33 |
| **Current diagnosis** | *n* MDD | 22 | 13 | | | 2.31 | .13 |
| | *n* ANX | 22 | 12 | | | 2.94 | .09 |
| | *n* CDA | 13 | 13 | | | .00 | 1 |
| | *n* HC | 40 | 10 | | | 18.00 | .00 |
| **Lifetime diagnosis** | % MDD | 43.29 | 72.92 | | | 11.31 | .00 |
| | % ANX | 41.24 | 60.42 | | | 4.74 | .03 |
| **MADRS** | Mean (SEM) | 7.10 (.94) | 12.30 (0.16) | | 1428 | | .00 |
| **BAI** | Mean (SEM) | 8.79 (1.04) | 11.91 (1.41) | | 1603 | | .01 |
| **Scan location** | % A/ L/ G | 28.9/ 41.2/ 29.9 | 37.5/ 39.6/ 22.9 | | | 1.34 | .51 |
| **Frequency of CEM** | %S/R/O/V | 0 | 8.5/ 51.0/ 21.3/ 19,2 | | | | |
| **Gray Matter** | Mean (SEM) | 740.40 (7.98) | 739.73 (8.79) | .00 | | | .96 |
| **White matter** | Mean (SEM) | 491.33 (6.94) | 499.57 (9.57) | .47 | | | .59 |

*Note*. CEM= Childhood Emotional Maltreatment, ANX= Anxiety Disorder, MDD= Major Depressive Disorder, CDA = Comorbid Depression and Anxiety Disorder, S=sometimes, R=regularly, O=often, V= very Often, A= Amsterdam Medical Center, L= Leiden University Medical Center, G= University Medical Center Groningen, MADRS= Montgomery Åsberg Depression Rating Scale, BAI= Beck Anxiety Inventory, F,U, $X^2$ = F ratio, Mann Whitney U statistic, and Chi-square test statistic, SEM= Standard Error of Mean.