Brain Activation in PTSD in Response to Trauma-Related Stimuli

Israel Liberzon, Stephan F. Taylor, Richard Amdur, Tara D. Jung, Kenneth R. Chamberlain, Satoshi Minoshima, Robert A. Koeppe, and Lorraine M. Fig

Background: *Repetitive recall of traumatic memories and chronic intermittent hyperarousal are characteristic of posttraumatic stress disorder (PTSD). Hyperarousal and memory dysfunction implicates "limbic" brain regions, including the amygdaloid complex, hippocampal formation, and limbic cortex, such as the orbitofrontal and anterior cingulate areas. To investigate the neurobiologic role of these brain regions in PTSD, we measured regional cerebral blood flow in PTSD with single photon emission computerized tomography (SPECT) during a symptom provocation paradigm.*

Methods: *Fourteen Vietnam veterans with PTSD, 11 combat control subjects, and 14 normal control subjects were studied with [^{99m}Tc]HMPAO in two sessions 48 hours apart: one session after exposure to white noise and the other following exposure to combat sounds. Skin conductance, heart rate, and subjective experience were recorded at the time of the studies.*

Results: *Activation for all three groups occurred in the anterior cingulate/middle prefrontal gyrus. Activation in the region of the left amygdala/nucleus accumbens was found in PTSD patients only. Deactivation was found in all three groups in the left retrosplenial region.*

Conclusions: *These findings implicate regions of the "limbic" brain, which may mediate the response to aversive stimuli in healthy individuals and in patients suffering from PTSD.* Biol Psychiatry 1999;45:817–826

Key Words: Neuroanatomy, rCBF, amygdala, limbic, combat

Introduction

Neurobiologic research in posttraumatic stress disorder (PTSD) has been focused on the aberrant regulation of the sympatoadrenal system and the hypothalamo–pituitary-adrenal (HPA) axis (Kosten et al 1987; Murburg 1994; Yehuda et al 1993). While much has been learned about neuroendocrine and psychophysiologic aspects of the stress response in this condition, the neuroanatomic underpinning of PTSD symptomatology remains unknown. The characteristic symptoms of PTSD, such as intrusive memories, recurrent dreams, and "flashbacks" of the traumatic event (American Psychiatric Association 1987) suggest abnormalities in the processing of memory—memory associated with traumatic, emotional material. Thus, investigations of the neurobiology of PTSD should logically seek to understand the neuroanatomic regions involved in memory and emotional regulation, in addition to the interaction of these systems with the CNS stress response.

The so-called "limbic" regions of the brain have long been associated with both memory and emotion. A broad concept, the limbic brain generally refers to amygdala, hippocampal formation, hypothalamus, thalamus, and nearby "paralimbic" cortex, such as the anterior cingulate cortex, orbitofrontal cortex insula, and temporal poles (Mesulam 1985; Nauta and Domesick 1982). Given the role of aberrant, intrusive, emotional memory in PTSD symptomatology, the limbic brain defines an obvious target of investigation; however, evidence linking these structures to specific PTSD symptoms is just emerging. Recent findings of structural abnormalities in the hippocampus of PTSD patients (Bremner et al 1995; Bremner et al 1997b; Gurvits et al 1996) support a strategy of investigating the functional integrity of these brain regions in PTSD.

Symptom provocation paradigms in combination with functional neuroimaging provide an important tool to visualize neuroanatomic correlates of psychiatric symptoms. Experimentally controlled exposure to trauma-related stimuli elicits exaggerated autonomic responses in

From the Psychiatry Service, Ann Arbor VAMC (IL, RA, TDJ, KRC); Nuclear Medicine Service, Ann Arbor VAMC (LMF); Department of Psychiatry, University of Michigan, (IL, SFT); Division of Nuclear Medicine, Department of Internal Medicine, University of Michigan, (SM, RAK, LMF) Ann Arbor, MI.

Address reprint requests to Israel Liberzon, MD, Psychiatry/PCT(116A), 2215 Fuller Rd, Ann Arbor, MI 48105.

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PTSD (Blanchard et al 1991; Pitman 1987). Similarly, by studying regional cerebral blood flow (rCBF) in PTSD patients during exposure to a trauma-specific narrative and a narrative describing neutral events, Rauch and colleagues found activation in the right amygdala, anterior cingulate cortex, and several other cortical areas (Rauch et al 1996). However, in the absence of a control group, the specificity of these findings to PTSD could not be ascertained. In a somewhat different imagery paradigm, Shin and co-workers found activation in the right amygdala of 7 PTSD patients (Shin et al 1997), but only during the mental imagery and not during the perception of combat images. As Shin and colleagues pointed out, these results are difficult to interpret. They also reported that during imagery of combat material, PTSD subjects activated the ventral anterior cingulate cortex (ACC) whereas control subjects recruited the superior ACC, relative to perception of combat material. Therefore, while preliminary findings support the involvement of limbic structures in PTSD symptom generation, questions remain about the extent, the diagnostic specificity, and the functional role of these findings.

To address some of these questions, we performed single photon emission computerized tomography (SPECT) scanning of PTSD patients and two control groups to measure regional cerebral perfusion in two settings: 1) immediately after exposure to a provocative stimulus; and 2) after a nonprovocative, control stimulus. As the provocative stimulus, we used combat sounds (e.g., helicopter sounds, small arm fire, etc.) which allowed the generation of images from the patient's own experience. For the nonprovocative stimulus, we presented the subjects with simple, white noise. During both conditions, we also simultaneously recorded psychophysiologic responses. Given the functional role of limbic and paralimbic cortex, we tested the hypothesis that the PTSD patients would show excitation of these regions in response to traumatic stimuli.

Methods and Materials

Subjects

Thirty nine subjects were recruited using advertisements within the hospital, local newspapers, churches, and veterans magazines. Fourteen Vietnam veterans with posttraumatic stress disorder, 11 Vietnam veterans without PTSD (combat controls), and 14 nonveteran male control subjects participated in this study (Table 1). The diagnosis of PTSD was established according to DSM-III-R criteria using the Structured Clinical Interview for DSM-III (SCID; Spitzer and et al 1990), administered by a trained clinician. Subjects with a history of psychotic disorders or dementia were excluded. Subjects with histories of substance abuse were in remission for at least 6 months. All subjects were right-handed and free of psychotropic medication for at least 4 weeks prior to the study (6 weeks in the case of fluoxetine). Control subjects were administered the nonpatient form of the SCID to exclude any active Axis I condition (schizophrenia or other psychotic disorders, affective disorders, anxiety disorders, dementia, substance abuse, or dependence). Combat control subjects had no past history of PTSD. Head CT scans were performed prior to the study to exclude structural abnormalities. All subjects gave written informed consent to participate in the study, which was approved by the Ann Arbor VAMC subcommittee on human subjects.

Table 1. Subject Characteristics

	PTSD Subjects (n = 11)	Normal Control Subjects (n = 14)	Combat Control Subjects (n = 11)
Mean Age (SD)	46.9 (.89)	45.4 (6.1)	51.2 (1.7)
Ethnicity			
white	10	11	9
black	1	2	2
Hispanic	0	1	0
Marital Status			
divorced/single	2	6	2
married	9	8	9
Comorbid Diagnoses			
MDD[a]	5	0	0
panic disorder	3	0	1
substance abuse/dependence[a]	7	5	1
PTSD symptoms			
impact of events	48 (2)	7 (10)	11 (4)
Mississippi	133 (5)	63 (4)	67 (4)
MMPI–PTSD	33 (5)	9 (1)	7 (2)
DES	24 (6)	6 (2)	12 (5)
SUDS	49 (10)	26 (5)	19 (7)

[a]In remission.

MDD, Major Depressive Disorders; DES, Dissociative Experiences Scale; SUDS, Subjective Units of Distress Scale (at baseline).

Procedure

Subjects were studied in two sessions on separate days, 48 hours apart, in counter-balanced order. One session consisted of exposure to nonspecific arousing stimuli (white noise), and the other to trauma-related stimuli (combat sounds). After securing intravenous access and electrophysiologic "hook-up," subjects were allowed a 30 min adaptation period. Physiologic indices of heart rate and skin conductance were collected during this adaptation period and throughout the duration of the session. Subjective distress pre- and post-stimulus administration was measured on 100 mm subjective units of distress scale (SUDS).

For the activation scan, a 3-min audiotape of combat sounds (e.g., helicopter sounds, explosions, and small arms fire) at gradually increasing volume (up to 75 dB) was played, according to the method of Blanchard and colleagues (Blanchard et al 1982). For the baseline scan, an audiotape of white noise with the same frequency spectrum was played at the identical, ramped intensity. The subjects closed their eyes to facilitate imagery during the specific uptake phase. Although some groups reported

greater variability of blood flow when subjects close their eyes, these findings occur when mental activity is not constrained by a particular task; furthermore, work from our center shows no significant difference in the coefficient of variation of occipital blood flow (Cameron et al 1990). Following the termination of the auditory stimuli, subjects were instructed to remain seated and to continue to imagine whatever came to mind for approximately 5 additional min. This 5 min period covered the uptake phase of the radioligand in the brain (Lassen et al 1988; Murase et al 1992).

Psychophysiologic Measures and Recording Equipment

Psychophysiologic data were recorded using an I-330 Interface System (J & J Enterprises, Poulsbo, WA). Skin conductance (SC) was recorded using Ag/AgCl electrodes attached to the second digit of the first and third fingers of the nondominant hand. These were connected to a T-601 Electrodermograph Module of the I-330 System, which utilized a constant voltage procedure (0.166 volts DC), with a range of 0 to 50 micromhos. Heart rate (HR) was recorded with a photoplethysmograph attached to the second digit of the second finger of the nondominant hand, connected to a P-401 Plethysmograph Module. Interpeak interval data was converted to beats per min, with a range of 40 to 200 bpm. Heart rate and skin conductance were sampled at 17 Hz. An IBM-compatible microcomputer running USE software (J & J Enterprises) controlled the data acquisition. Mean electrophysiologic scores were computed for a baseline period prior to the onset of audiotaped sounds (PRE; duration = 120 sec), and imagery period following termination of audiotaped sounds (duration = 150 sec).

Brain Scintigraphy—Acquisition

For each SPECT study, subjects were injected intravenously with 30 mCi [^{99m}Tc]HMPAO, 30 sec prior to the end of the auditory stimulus. Since psychophysiologic data indicate that in PTSD patients, the effects of the traumatic stimulus persist for several minutes (Pitman 1987), we timed the arrival of the [^{99m}Tc]HMPAO to the brain to correspond with termination of the auditory stimulus. To minimize extracranial blood flow to the temporal muscles from teeth clenching, subjects were instructed to keep their mouths open during injection and for 10 minutes thereafter, as suggested by Drevets and colleagues (Drevets et al 1992). SPECT acquisition began approximately 60 min after tracer injection to maximize brain-to-soft tissue ratios. A Siemens MultiSPECT 3 triple-headed gamma camera with low-energy, high-resolution collimation was used for the first four PTSD patients acquired. Subsequent PTSD patients and all control subjects were imaged on an ADAC Genesys Vertex dual-head camera using ultrahigh-resolution collimators. We adjusted the acquisition duration to match the total number of counts acquired for the different systems. Technical performance parameters, such as spatial resolution (between 8 and 9 mm for both systems) and image uniformity, as well as the collimators employed for the studies were very similar between cameras, indicating that the data could be pooled. Most importantly, however, since each subject serves as his own control in the subtraction analysis, small differences in performance of the imaging devices are far less important sources of variation than the biologic differences. SPECT data were collected from 64 projections and reconstructed into a 128 × 128 image matrix. Each projection was acquired for 30 sec and a Butterworth filter (cutoff 0.2/order 5.0) with analytic y-axis filtering and Chang's attenuation correction (mu value for attenuation coefficient = 0.15) was employed for image reconstruction to full width half, maxima of 9.5 mm.

Image Analysis

Data analysis was performed using fully automated routines developed at the University of Michigan for analysis of PET activation studies (Minoshima et al 1993a) and adapted for SPECT image data. This image registration method has been previously validated with brain SPECT images (Bartenstein et al 1997). Others have also demonstrated that methods originally developed for PET and fMRI studies can be successfully applied to SPECT activation data (Matthew and Hill 1998). The procedure involves the following steps.

1. Intrasubject image registration: The two image sets obtained from the same subject on different days were realigned to a common orientation using an automated computer algorithm (Minoshima et al 1992). During this coregistration step, each image was also normalized to uniform whole brain activity.
2. Anatomic standardization: All image sets were transformed to a standard stereotactic coordinate system using an algorithm based upon detection of the midsagittal plane and the line passing through the anterior and posterior commissures (Minoshima et al 1993b). This was followed by anatomic standardization via linear scaling and nonlinear shape deformation (Minoshima et al 1994), smoothing with a Gaussian kernal (9 mm at full width, half maximum) and finally voxel-by-voxel analysis across multiple subjects.
3. Image averaging: A "difference" image set (combat sounds vs white noise) was created for each subject, and then the subtraction image data were averaged across multiple subjects for each subject group. Using the intersubject variance averaged across all analyzed voxels (pooled variance), mean differences in CBF were converted to *Z*-statistic maps (Worsley et al 1992).
4. A priori regions: We identified an a priori search area in parts of the limbic brain and paralimbic cortex thought to be important for memory and regulation of anxiety, based on findings available at the initiation of the study (Baxter et al 1987; Baxter et al 1988; George et al 1995; Pardo et al 1993). This included subcortical limbic structures of the medial temporal lobes (amygdala and hippocampus) and paralimbic cortex (parahippocampal gyrus, orbitofrontal cortex, and medial temporal poles). Using the Talairach and Tournoux atlas coordinates, we defined the medial temporal lobe region-of-interest as beginning 20 mm anterior to and 10 mm inferior to the anterior commissure, and terminating 40 mm posterior to the anterior commis-

sures. We defined an anterior frontal region, including orbitofrontal cortex and inferior medial frontal cortex, as anterior to the anterior commissure 10 mm, and 10 mm inferior to the bicommissural line. Within these areas, we accepted as significant any focus with a maximum voxel $Z > 2.6$, corresponded to an uncorrected significance level of $p < 0.005$.

5. Image-wide search: To explore brain regions outside of the a priori regions-of-interest and to identify common areas of activation across all three experimental groups, we searched the averaged difference image for each group images and a composite averaged-image of all three groups. We calculated the corrected probability threshold for activation, accounting for the effective number of comparisons performed over the entire image set. With this procedure, a Z score of approximately 4.4 corresponded to a false-positive expectation of 0.05 (Friston et al 1991; Worsley et al 1992); and we report statistical trends with $Z > 4.0$.
6. Testing for group differences: We localized the atlas coordinates of foci in the image for the combined group and then centered a spherical volume of interest on those coordinates in the anatomically standardized images of each subject. Using a simple paired t test, we tested for a significant difference ($p < 0.05$) between the white noise and combat sounds conditions. Because of reported structural differences in the medial temporal lobes of patients with PTSD, we chose not to use a voxel-by-voxel group comparison of warped, anatomically standardized images. It is the experience of our lab that warping may not perform unbiased adjustments where group structural differences exist.

Results

Psychophysiology and Subjective Distress

PTSD subjects had significantly higher heart rate (repeated measure ANOVA effect of group, F [2,28] = 3.48, $p < 0.05$), higher skin conductance (trend level, F [2,31] = 2.95, $p < 0.07$), and subjective distress (F [2,28] = 2.97, $p < 0.06$), as compared to either control group. There was also a significant main effect of condition (combat sounds vs white noise) on skin conductance (F [1,31] = 18.8, $p < 0.001$) and subjective distress (F [1,31] = 36.8, $p < 0.001$). PTSD patients had significantly larger subjective distress and skin conductance responses to combat sounds relative to white noise, compared to either control group (group by condition interaction, F [2,31] = 6.16, $p < 0.006$ and F [2,31] = 3.35, $p < 0.05$, for SUDS and SC respectively). Mean SUDS and skin conductance responses are presented in Figures 1 and 2. There was no significant difference between the two control group responses in either distress levels or psychophysiologic measures. The observed HR response occurred in the same direction as SC; however, the between-group differences did not reach statistical significance (data not shown).

Cerebral Blood Flow

One of our PTSD subjects experienced a full-blown "flashback" during the presentation of the combat sounds. He exhibited an unusual pattern of rCBF, with a dramatic

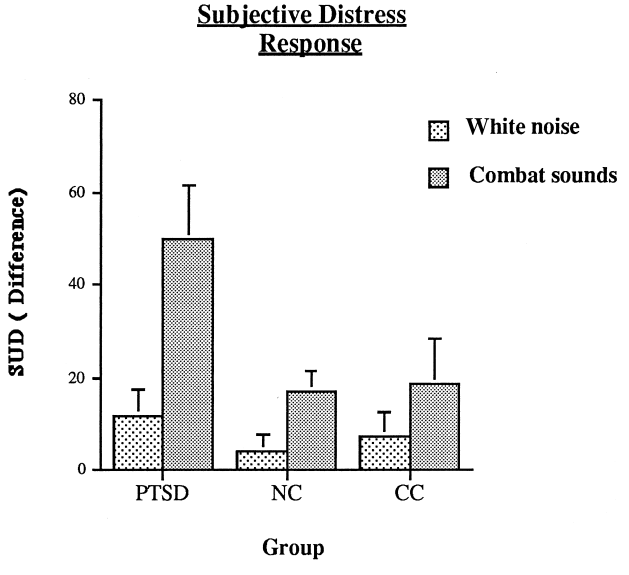

Figure 1. The change in subjective distress (SUD—subjective units of distress) in response to white noise and to combat sounds in PTSD patients, combat controls and normal controls. Baseline score is subtracted from the post-stimulus score.

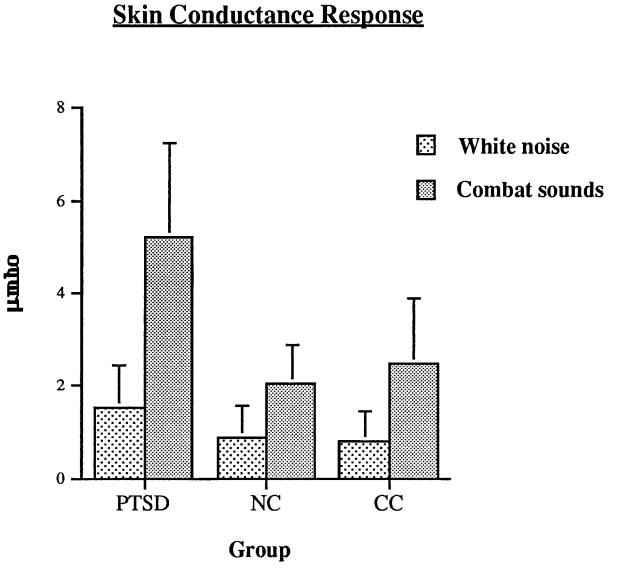

Figure 2. Skin conductance response to white noise and combat sounds in PTSD patients, combat control subjects, and normal control subjects. The baseline measure is subtracted from the averaged skin conductance during the imaging period.

Table 2. Regional CBF Peaks and Their Coordinates

Region	PTSD Patients		Combat Control Subjects		Healthy Control Subjects		Composite Image	
	(x, y, z)[a]	*Z* Score[b]	(x, y, z)[a]	*Z* Score[b]	(x, y, z)[a]	*Z* Score[b]	(x, y, z)[a]	*Z* Score
Activation within a priori regions								
Left amygdaloid region	(−21, 3, −9)	2.65	—	—	—	—	—	—
Activation outside a priori regions								
Relative increase								
Anterior cingulate/Medial	(3, 46, 11)	2.71	(6, 48, 2)	2.66	(1, 50, 0)	3.12	(3, 48, 2)	4.18
prefrontal cortex	(6, 68, −4)	3.18						
Relative decrease								
Right retrosplenial region	(15, −49, −9)	−4.44	(15, −58, −2)	−2.79	(6, −49, −9)	−2.87	(15, −49, −7)	−4.45

[a]Stereotactic coordinates from Talairach and Tournoux atlas (1988), left/right, anterior/posterior and superior/inferior, respectively.

[b]Foci listed as significant if: $Z > 2.6$ (uncorrected $p < 0.005$) in a priori regions, or *Z* score > 4.4 ($p < 0.05$, corrected for image wide search) or *Z* score $Z > 4.0$ (trend level). Sub-threshold activation foci in brackets are listed for purposes of comparison when a supra-threshold focus appeared in another group or in the composite image.

alteration in cortical relative to subcortical flow. This pattern was not consistent with the global depression in CBF that one might observe during hyperventilation. Because of the unusual experience of this subject and the extremely different CBF pattern for this scan compared to all other scans, we omitted his data from this analysis and reported his results separately (Liberzon et al 1997). The scans from two other subjects could not be effectively analyzed together with the rest of the scans, because of tracer uptake in extracerebral tissue, creating image distortion and rim artifacts. Therefore, these subjects' data (both scans and psychophysiology) were excluded from the analysis.

Activation Within A Priori Defined Regions

The only activation foci exceeding statistical thresholds in the a priori limbic area occurred in the PTSD group (Table 2). Figure 3 shows patterns of differential rCBF in all three groups, which included a focus in the region of the left amygdala, extending into the nucleus accumbens of the PTSD group. Because of the limited resolution of this imaging modality and the neuroanatomic proximity of small structures in this area, sometimes referred to as "extended amygdala" (Alheid and Heimer 1988; Paxinos 1990), we referred to this focus in general terms as the "amygdaloid region," without implying a more specific

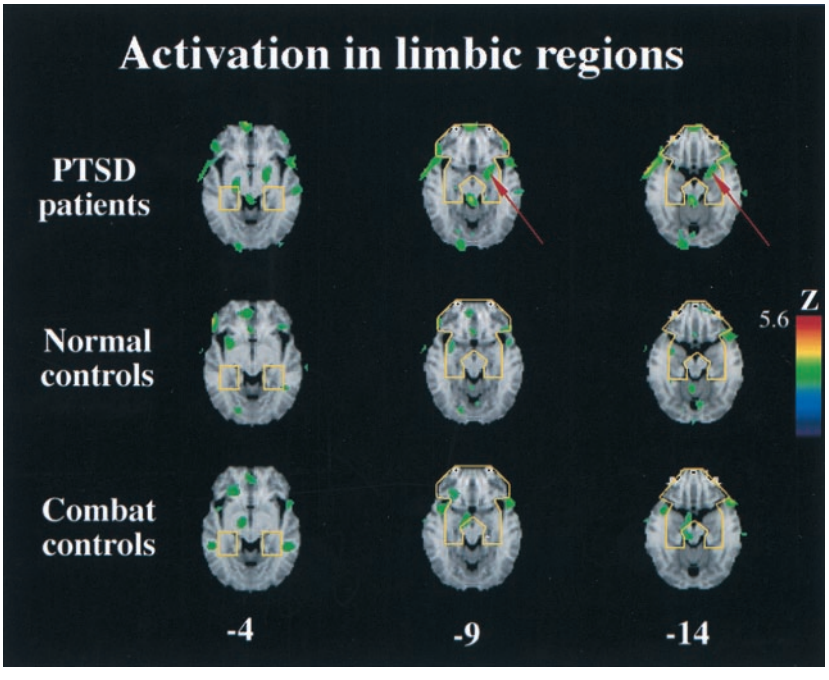

Figure 3. The image depicts relative activation (combat sounds compared to white noise) in limbic regions in PTSD patients, and combat and normal control subjects. The a priori region of interest is indicated by the yellow border. The images show Z-values > 1.65 (uncorrected) superimposed on a reference MRI image in atlas space and in radiological orientation (left/right reversed). The level below the bicommissural line, in mm, is indicated beneath the bottom row of images.

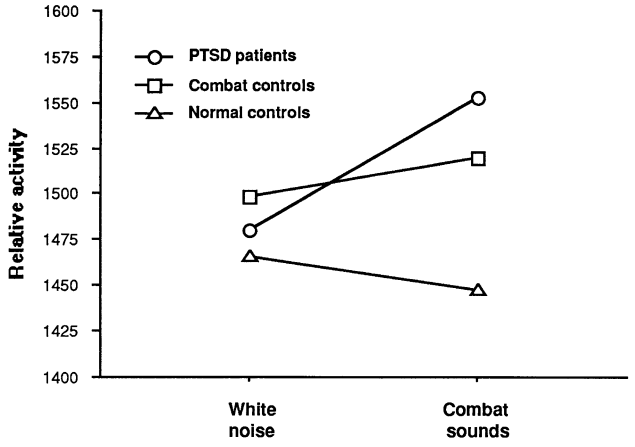

Figure 4. Activity in the region of the left amygdala was approximately equal in all three groups during the presentation of white noise and then rose sharply during combat sounds, only for the PTSD group. Relative activity was measured at a spherical, 18 mm VOI centered on the coordinates of the peak present in PTSD group.

structure. For illustrative purposes, Figure 4 shows relative rCBF for each group at a spherical VOI centered on the coordinates of the PTSD group. Activity during the white noise condition is approximately identical in all three groups, but rises sharply in the PTSD group only. Although no comparable activation appeared above threshold in this region in either of the two control groups, the VOI analysis (18 mm diameter sphere) of this focus, using stereotactic coordinates from the composite image, did not show significant differences in activation between the groups. We did not identify any supra-threshold foci for any group along the length of the parahippocampal formation and overlying gyrus.

Activation Peaks from Image-wide Search

In the image-wide search, we found a trend-level activation focus in the anterior cingulate/medial prefrontal cortex for the composite image. Examination of the averaged image for each group separately revealed similar activation foci in this region, at Z values less then our image-wide threshold ($Z = 2.6$ to 3.12; see Table 2 and Figure 5). In addition, we found activation peaks in the vicinity of the right and left temporal poles in all three groups of subjects. Activation in the left temporal pole appeared larger and more dorsal in the PTSD patients, as compared groups to combat and normal control groups. However, the local maxima for all of these activations mapped to outside of brain tissue (x, y, z coordinate foci: 60, 8, −20 and −62, 14, 2 for PTSD group; 66, −1, −16 and −51, 21, −18 for control groups), close to the location reported by other groups for teeth clenching. (Drevets et al 1992; Rauch et al 1996). Thus, we interpreted these activation foci adjacent to the temporal poles as likely artifacts due to contraction of the temporalis muscle and other extracerebral muscle activity.

Relative Deactivation

We found a significant "deactivation," where normalized flow was less in the combat sounds condition compared to the white noise condition, in the right retrosplenial region in all three subject groups. The magnitude of the deactivation in the PTSD group appeared larger than that observed in the combat or normal control groups ($Z = 4.40$ compared to 2.79 and 2.87, respectively; Table 2).

Correlation

We examined potential relationships between psychophysiologic responses, subject ratings, and cerebral perfusion by calculating Pearson correlation coefficients for rCBF changes and changes in heart rate, skin conductance, and SUDS. Regional CBF changes were calculated within volumes of interest centered on the activation and deactivation peaks. No correlation with $p < 0.01$ was identified between rCBF change and these ancillary measures, neither when all the subjects were examined together nor when groups were analyzed separately.

Discussion

During exposure to traumatic stimuli, the PTSD patients demonstrated the characteristic exaggerated psychological and psychophysiologic responses, similar to those reported in the literature (Pitman 1987; Pitman et al 1990). We also found that the PTSD patients increased rCBF in relevant areas of the limbic brain in the vicinity of the left amygdala/nucleus accumbens. No comparable activation was found in either control group. All three groups exhibited a response in the anterior cingulate/medial prefrontal cortex, suggesting that at least some of the response to the potent auditory stimuli is not specific to PTSD. Given the role these neuroanatomic structures play in the regulation of mental processes important to PTSD, our findings support and extend the existing evidence of limbic system and extended amygdala involvement in PTSD symptomatology.

Activation in the region of the extended amygdala in response to traumatic stimuli is entirely consistent with the role of this area in emotion. Of the variety of emotional responses which implicate the amygdala, fear and avoidance behavior stand out prominently (Davis 1992b; LeDoux 1992). As a part of the limbic brain, it forms one part of a network including the hypothalamus, septal nuclei, hippocampal formation, substantia innominata, midbrain, nucleus accumbens, and medial thalamic nuclei (Alheid

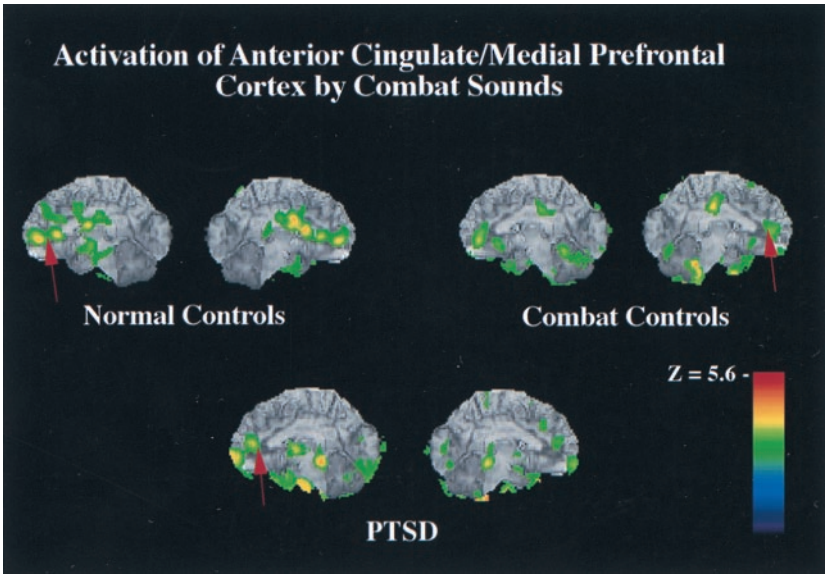

Figure 5. Activation peaks projected to the medial aspect of the cortex show activation in the anterior cingulate/medial prefrontal cortex of all three groups. Display conventions are as in Figure 3.

and Heimer 1988; Paxinos 1990). With the limited resolution of our imaging technique, we cannot distinguish activation in the nucleus accumbens from an adjacent activation in a medial forebrain structure such as the substantia innominata, but it is clear that the activation focus shown in Figure 3 falls well within the borders of this functional network. Relevant for PTSD, the amygdala and associated structures modulate memory acquisition of salient stimuli, particularly with aversive content. In animal models of fear-conditioned responses, lesions of the amygdala block the acquisition of conditioned responses to fear-associated stimuli (Adolphs et al 1995; Davis 1992a; LeDoux 1992). Finally, recent functional neuroimaging studies in humans have identified activation of the extended amygdala in response to affective visual stimuli (Taylor et al 1998; Breiter et al 1996; Reiman et al 1997).

Our results also partially replicate and extend prior functional neuroimaging work with PTSD patients. As observed in the study by Rauch and co-workers, (Rauch et al 1996), we found activation of the amygdaloid region during imagery, even though Rauch and co-workers used personalized scripts to induce imagery, while we used generic combat sounds. In contrast to their study, the present study employed two distinct control groups, neither of which exhibited activation patterns similar to that shown by the patient group, suggesting that this activation pattern might be specific to PTSD. Shin and colleagues also found activation in the amygdaloid region of PTSD patients and not in their control group (Shin et al 1997). However, no activation was detected during the presentation of traumatic material, which complicates the interpretation of their finding as an activation associated with emotion. Both earlier studies found activation in PTSD in the right amygdala, while we observed activation on the left. Future studies will be needed to explore the functional significance of this laterality.

While our results may suggest involvement of the amygdaloid region in the generation of PTSD symptoms, alternative interpretations should be considered. Perhaps most importantly, a failure to demonstrate an activation focus is not the same as the demonstration of the absence of activation. We cannot rule out the possibility that our control subjects did recruit limbic structures in response to the combat stimuli. However, the difficulty of demonstrating group differences in image-averaged, statistical parametric maps prevented us from showing a statistically significant difference between groups. We would expect that larger sample sizes should eventually demonstrate specific patterns of activation in PTSD subjects.

Another issue arises with the confounding of emotional content and episodic personal memory. For instance, evocation of meaningful personal memory in general, not specifically associated with PTSD, might have produced the activity in the region of the left amygdala. To distinguish these possibilities, the presence of a control group with a similar personal experience, i.e., exposure to combat, is needed. Thus, the absence of activation in limbic regions in both control groups is consistent with the conclusion that the activation of the amygdaloid region in PTSD patients could be a specific response. Alternatively, one might interpret this activation as reflecting a more intense emotional response in the PTSD patients. As the psychophysiologic data indicated, the PTSD patients exhibited greater arousal, and this could have caused the activation in the amygdaloid region, unrelated to the evocation of traumatic memory. In PET studies of normal control subjects exposed to strong negative affect (Taylor et al 1998b), we did find activation of the left amygdaloid

region in the initial but not in subsequent exposures to negative stimuli. We interpreted this as evidence of a habituation process. If this interpretation is accurate, the activation we found in the amygdaloid region of PTSD subjects might reflect their "failure to habituate" to trauma-related cues. From this experiment alone, we cannot conclude whether the amygdaloid activation represents the response to a more intense experience of the PTSD patients or a causal agent of the disorder.

Activation in the ACC (plus medial prefrontal cortex) was found in all three groups of subjects. These findings are consistent with the role of ACC in emotional processing, which exchanges projections with medial thalamic nuclei (Alexander et al 1986) and has significant connections with the amygdala (Devinsky et al 1995; Mesulam 1985). Considered a part of the paralimbic cortex, the ACC is thought to play a critical role in the assignment of motivational significance. ACC activation has been demonstrated in tasks requiring the recognition of facial emotion (Bremner et al 1997a), self-induced dysphoria (George et al 1995), phobic anxiety (Rauch et al 1995), and obsessive compulsive symptom provocation (Rauch et al 1994). Previous neuroimaging studies in PTSD patients have reported anterior cingulate activation (Rauch et al 1996; Shin et al 1997), although under different conditions. It is possible that ACC activation is associated with a "nonspecific" anxiety state. The fact that both control groups found combat sounds disturbing (evident from the increase in SUDS ratings) further supports this interpretation.

We interpreted activation located on the edge of the temporal poles as reflecting extracranial muscle activity during the active scan condition. While it is possible that extracranial activity masked true activation foci in the temporal poles, we suggest the more cautious interpretation. The bilateral appearance, the coordinates of local maxima falling outside brain tissue, and the prior work of other laboratories with teeth clenching paradigms build a strong case that these peaks represent extracerebral artifact. During symptom provocation paradigms, PTSD patients showed greater activity of the facial muscles while experiencing stimuli relevant to their trauma, compared to neutral conditions and to normal control subjects (Blanchard et al 1991; Pitman 1987), a fact which could explain the larger foci observed in the PTSD group. Even our control subjects reported more arousal during the active condition, congruent with a larger skin conductance response. Although our subjects held their mouths open as instructed, this tactic apparently did not ensure the absence of increased muscle tension during the more anxiety-provoking exposure to combat sounds.

We did not find any significant correlations between the blood flow changes at the activation peaks and the observed psychophysiologic changes. This is not entirely surprising since psychophysiologic measures often do not correlate well with other neurobiologic responses, such as catecholamine secretion or HPA axis activity (Malarkey et al 1995). It is also possible that a larger sample size is needed to obtain a significant correlation for rCBF and psychophysiologic responses.

Our subjects exhibited a relative decrease in CBF in the right retrosplenial region. The significance of these deactivations is not entirely clear. The larger relative decrease for the PTSD group could reflect traumatic experience inducing a true "deactivation" in these regions, but we cannot determine from this data set alone whether this represented rCBF decrease during the combat sounds condition or CBF increase during the white noise condition. The retrosplenial and parietal regions have been frequently activated during recall of episodic memories, along with the precuneus (Andreasen et al 1995; Fletcher et al 1995; Kapur et al 1995), and we did find a relative decrease in the precuneus, but with *Z* scores just below our cutoff threshold. While it may seem counterintuitive that recall would occur during the condition without the provocative stimulus, the absence of differentiated stimuli may have caused our subjects to recall more diverse experiences. However, the speculative nature of these interpretations will require additional studies to test the competing hypotheses, e.g., comparison with a third, "neutral" task condition.

The methodology of this study entails certain limitations. Measurement of cerebral perfusion was done qualitatively and normalized for uniform global activity. Global decreases in CBF during acute anxiety have been reported in the literature (Mountz et al 1989; Stewart et al 1988), results usually attributed to hyperventilation and resulting hypocapnia (Mathew and Wilson 1990). In the absence of quantitative protocol for measurement of CBF, the [^{99m}Tc]HMPAO SPECT technique, as typically applied, will not detect global CBF changes. However, these could not have induced the regionally specific rCBF changes we observed. Another potential confound was that four subjects in this study were imaged with a Siemens camera and the remaining 32 with an ADEC camera. While this could contribute additional variability to the data, the main analysis in this study involved within-subjects analysis of difference images. In this sense, each subject served as his own control, and camera-specific contributions to the change measures are canceled out in the subtraction. Nevertheless, we also analyzed the data excluding the four PTSD subjects who were imaged with the Siemens camera. This did not change our results, except for lower degrees of freedom and less robust statistical significance (as expected with fewer subjects). Therefore, while some limitations exist in use of two

cameras, they would not appear to explain or seriously confound our results.

In summary, our findings suggest that activation patterns seen in PTSD involve a number of functionally diverse regions. Activation of the ACC/medial prefrontal area may mediate a generalized, nonspecific response to emotional experiences. Activation of the extended amygdala/medial forebrain region may reflect emotional responses specific to PTSD pathophysiology, but the failure to find this activation in our control groups does not yet establish a finding specific to PTSD. Finally, the relative decreases that we observed in all three groups might suggest that some emotional responses are reflected both by activation of certain brain regions and deactivation of others. These interpretations are necessarily preliminary and additional data are needed to exclude alternative hypotheses. However, the data do identify neuroanatomic regions where future work might examine the emotional response to stressful stimuli and the phenomena of PTSD.

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