Smaller right hippocampus in war veterans with posttraumatic stress disorder

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Received 17 January 2006; received in revised form 14 June 2006; accepted 27 August 2006

Abstract

Chronic stress can putatively cause damage in the human hippocampus, but evidence of damage has not been consistently shown in studies on hippocampal morphology in posttraumatic stress disorder (PTSD). We compared hippocampal volumes in PTSD patients and normal subjects. Using a 3D T1-weighted GRE magnetic resonance imaging sequence, we measured hippocampal volumes in 15 war veterans with combat-related chronic PTSD and 15 case-matched normal controls. Although war veterans, our PTSD subjects were not professional soldiers and were mobilized shortly before they were exposed to a very specific combat-related trauma over a 3-day period. In our study, the period between traumatic exposure and imaging was considerably shorter, on average, 9 years, compared with at least two decades in previous studies on subjects with combat-related PTSD. Moreover, our subjects were free of any comorbidity, treatment or medication. The right hippocampus was significantly smaller in PTSD subjects than in controls. The left hippocampus was also smaller in PTSD subjects than in controls, but the difference was not significant. In all PTSD subjects, the right hippocampus was smaller than the left (on average, 7.88%). Our results show smaller volume of the right hippocampus in PTSD patients than in normal subjects.

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Keywords: Hippocampus; PTSD; War veterans; Laterality; MRI

1. Introduction

Posttraumatic stress disorder (PTSD) develops in some people after exposure to an extreme stressor or traumatic event (American Psychiatric Association, 1994). It is characterized by three distinct types of symptoms consisting of reexperiencing of the event, avoidance of reminders of the event, and hyperarousal, which continue for a substantial period of time. Reexperiencing of the event refers to unwanted recollections of the incident in the form of distressing images, nightmares or flashbacks. These symptoms indicate...
dysfunction in acquisition and extinction of fear conditioning, and the hippocampus has been the most consistently implicated brain region (McEwen, 2000; Yehuda, 2002).

Although hippocampal changes associated with PTSD have been extensively studied, there is currently little consensus regarding whether the hippocampus of patients with PTSD is, in fact, reduced in volume. Several studies reported smaller hippocampal volumes in combat veterans with PTSD (Bremner et al., 1995; Gilbertson et al., 2002; Gurvits et al., 1996), in adults with PTSD secondary to child abuse (Bremner et al., 1997), in female adult survivors of childhood sexual abuse (Stein et al., 1997; Bremner et al., 2003), in adults with different kinds of trauma, but mostly childhood sexual abuse (Vallarreal et al., 2002), and in traumatized police officers (Lindauer et al., 2004). Reductions in hippocampal volume ranged between 5% and 26%. Other studies did not confirm the finding of smaller hippocampal volume in trauma survivors from an emergency room (Schuff et al., 2001). In our study, the period between traumatic exposure and imaging was considerably shorter—on average, 9 years—compared with at least two decades in previous studies. In addition, our subjects were free of any comorbidity, treatment or medication linked to changes in hippocampal volume (Laakso et al., 2000; Frodl et al., 2002; Sheline et al., 2003; Vermetten et al., 2003).

2. Methods

2.1. Participants

During 2004, we screened 82 PTSD patients who came to our institution for the first time and who had not previously been treated. Fifteen subjects who fulfilled all the criteria described below were selected for this study. All the subjects gave their written informed consent to participate in the study. The study was approved by the ethical committee of the Clinical Center Zagreb, University of Zagreb, School of Medicine.

As part of the screening procedure, all the subjects underwent an individual psychiatric examination performed by two psychiatrists with more than 5 years of experience in work with psycho-traumatized patients. For evaluation of psychological distress attributable to war trauma exposure, all subjects underwent structured clinical interviews according to the clinical version of the International Classification of Diseases and the Diagnostic and Statistical Manual of Mental Disorders (World Health Organization, 1993; American Psychiatric Association, 1994). Interviews provided data on previous and comorbid psychic disturbances, heredity, traumatic experiences, current symptoms, duration of symptoms, intensity, and possible treatment.

In order for a subject to be enrolled in the study, he had to have a diagnosis of chronic PTSD, and any other lifetime neurological or psychiatric disorder had to be ruled out. Signs or symptoms of depression and/or anxiety were required to be secondary to PTSD and not related to a depressive or other anxiety disorder. The participants included had to be free of any history of lifetime alcohol abuse or dependence both in self-report and heteroanamnesis. Only the patients for whom both interviewers agreed regarding the PTSD diagnosis and the lack of comorbidity were enrolled in this study. The participants selected received no psychotropic medication for at least 6 months preceding the study.

Before the examination, a detailed heteroanamnesis was obtained from the families (spouse, children and parents) and subjects’ general practitioners. Every participant in the study had had the same general practitioner since he completed high school, as is customary in Croatia. The heteroanamnesis provided relevant medical (general health, alcohol and substance abuse histories) and sociodemographic data (e.g., education, qualification, employment and marital status, number of children, place of residence, data about previous and current disturbances, traumatic experiences, and previous and current social functioning). In the selection of PTSD
subjects, special attention was given to recruit those with adequate pre-traumatization general health, mental health, and social functioning. Only the subjects with no limitations in physical and social activities due to either physical or emotional problems, with no limitations in usual role activities, with no psychological distress, with adequate vitality and general health, as reported by themselves, their family members and their general practitioners, were included in the study.

As part of the screening procedure, a thorough physical and neurological examination was conducted, and none of the participants suffered from any comorbid neurological or physical conditions. All the participants had to have normal results on the Mini Mental State Examination (Folstein et al., 1975). As far as the subjects could remember, none of them ever suffered head trauma or loss of consciousness.

All the participants in the study had to be Croatian War veterans who actively participated in combat and were exposed to multiple combat-related traumatic experiences such as witnessing the death or wounding of fellow soldiers and/or civilians, and being exposed to a sudden artillery and/or other military attack. Their traumatic experiences occurred specifically during a major Croatian Army offensive that took place from August 4 to 7, 1995 (Lang et al., 1997). None of them were professional soldiers before or after this engagement. They were all mobilized a few months before the offensive and were demobilized shortly afterwards. They were not exposed to any combat activity before or after the offensive. After demobilization, they returned to their normal pre-mobilization life and activities. When they were drafted for active military service, the subjects passed rigorous entrance tests, which ensured that they did not have any prior abnormal personality trait or disorder. Unfortunately, these data were not available in greater detail from the military authorities for legal reasons.

Following the offensive, they developed PTSD symptoms, but requested no professional help until they were enrolled in this study. Due to the long period of time that elapsed from the traumatization, the exact dates of symptom onset were not possible to determine for any of our subjects. Nevertheless, they all reported that their symptoms had started during the first year following the offensive.

For each PTSD subject, a healthy control was case-matched for age, sex, handedness, years of education, and socioeconomic level. All participants, both PTSD and controls, were right-handed males. All control subjects were interviewed by the same psychiatrists as PTSD subjects. The same physical and neurological examination was conducted, and the same procedure was used to obtain relevant heteroanamnestic data.

The ages of the participants at the time they were enrolled in this study ranged from 34 to 53 (mean 41 ± 5.37). The ages of the PTSD subjects at the time of traumatization ranged from 24 to 41 (mean 32 ± 5.44), so the period between trauma exposure and imaging was on average 9 ± 0.49 years.

All participants, both PTSD patient and controls, had 8 years of elementary education and 4 years of high school. Socioeconomic level was determined from the auto- and heteroanamnesis on the basis of qualification, employment status, place of residence, and previous and current social functioning. All participants in both groups were of middle professional qualification as determined by categorization generally used in Croatia (low and high are two other possible classifications). All participants had steady jobs in their professional qualification with regular incomes. They were all from an urban population, and their social functioning (both previous and current) was considered normal and adequate by their families.

2.2. MRI data acquisition

The magnetic resonance studies were performed on a "Prestige Gyrex" GE/Elscint scanner with 2.0-Tesla field strength using a standard head coil. The magnetic resonance imaging (MRI) sequence used for volumetric analysis was a 3D T1-weighted GRE acquisition of the whole brain (repetition time = 540 ms, echo time = 20 ms, field of view = 240 × 240 mm, matrix = 256 × 192). Slices obtained were 1.1 mm thick without an interleave gap, with 1.0 × 1.0 mm inplane resolution, and oriented orthogonal to the long axis of the hippocampus.

2.3. MRI data processing

The boundaries of the hippocampus were outlined manually on each slice. Beginning rostrally, at the posterior uncus, the hippocampus was outlined with the superior border defined by the alveus, demarcating the amygdala superiorly from the underlying hippocampus, excluding the parahippocampal gyrus medially and its white matter inferiorly. Progressing posteriorly, its superior and medial border were defined by the temporal horn of the lateral ventricle, the lateral border by the transverse (choroid) fissure, and the inferior border by the hippocampal sulcus. When the hippocampal sulcus was not open, a line was drawn from its indentation to the temporal horn. The ammonic horn takes on its typical appearance at the level of the lateral geniculate through the
body of the hippocampus. Measurements at this level included the dentate gyrus, the cornu ammonis, the subiculum, and the alveus and fimbria. The limit between the subiculum and the transentorhinal cortex of the parahippocampal gyrus was defined by a line from the inferior border of the subiculum to the medial edge of the dentate gyrus as it curves into the hippocampal sulcus. The posterior boundary was the last coronal section before visualization of the crus fornix in its entire length and width as it ascended toward the thalamus. The hippocampal tail was not contoured. Head movements were corrected for along the anterior commissure–posterior commissure (AC-PC) line and the intrahemispheric fissure before measurement.

The surface of the hippocampal structure was measured on separate slices using DicomWorks v 1.3.5 software (2000–2001; Philippe Puech, Loic Boussel), and the volume was calculated by multiplying the surface area and the slice thickness. In order to compensate for head and brain volume differences among the subjects, we normalized the raw hippocampus volumes against the mid-sagittal intracranial surface, using Gullap’s formula (Yucel et al., 2002):

\[
\text{NHV} = \frac{\text{MMAG} \times \text{AHV}}{\text{MAS}}
\]

where \(\text{NHV}\) = normalized hippocampus volume; \(\text{MMAG}\) = mean mid-sagittal area of the group; \(\text{AHV}\) = absolute hippocampus volume; and \(\text{MAS}\) = mid-sagittal area of the subject (Fig. 1).

### Table 1

<table>
<thead>
<tr>
<th>Subjects</th>
<th>N</th>
<th>Mean</th>
<th>S.D.</th>
<th>(t)</th>
<th>(P^a)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mid-sagittal area (cm²)</td>
<td></td>
<td></td>
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<td></td>
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<tr>
<td>PTSD</td>
<td>15</td>
<td>152.86</td>
<td>10.581</td>
<td>0.671</td>
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<tr>
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<td>15</td>
<td>155.18</td>
<td>8.152</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Norm. Vol. H. r. (cm³)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PTSD</td>
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<td>4.07</td>
<td>0.513</td>
<td>3.267</td>
<td>≤0.05</td>
</tr>
<tr>
<td>Control</td>
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<td>4.62</td>
<td>0.623</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Norm. Vol. H. l. (cm³)</td>
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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PTSD</td>
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<td>4.39</td>
<td>0.537</td>
<td>0.485</td>
<td>&gt;0.05</td>
</tr>
<tr>
<td>Control</td>
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<td>4.44</td>
<td>0.562</td>
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<td></td>
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<td>Norm. Total H. Vol.</td>
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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PTSD</td>
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<td>8.326</td>
<td>1.073</td>
<td>−1.993</td>
<td>&lt;0.05</td>
</tr>
<tr>
<td>Control</td>
<td>15</td>
<td>9.11</td>
<td>1.085</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

\(^a\)Student’s \(t\)-test.

### 2.4. Statistical analysis

The measurement was performed by three neuroradiologists unaware of the patient’s diagnosis, the purpose of measurement, or each other’s results. Both hippocampi in each subject were measured by each examiner and three values were produced. The mean value for each variable was calculated, normalized against the mid-sagittal cranial surface, and used in statistical analysis.

Interclass correlation coefficients were calculated. Intra-rater reliabilities were 0.96 for rater no. 1; 0.96 for rater no. 2; and 0.97 for rater no. 3. Inter-rater reliabilities were very high as well. For the three combinations of rater couples, reliabilities were 0.92, 0.94 and 0.96 for the right hippocampus; and 0.91, 0.93, 0.91 for the left hippocampus.

Hippocampal volumes were compared with a two-tailed \(t\)-test. Statistical significance was set at 5% (\(\alpha=0.05\)).

### 3. Results

Table 1 presents mean values, standard deviations (S.D.), and other statistical values for the volumetric measurements. Subjects with PTSD had significantly smaller right hippocampi (13.5%) than normal control subjects (4.07±0.513 cm³ vs. 4.62±0.623 cm³, \(P\leq 0.05\)). The left hippocampus was also smaller in PTSD subjects (1.1%), but the difference was not significant (4.39±0.537 cm³ vs. 4.44±0.562 cm³, \(P>0.05\)). Total hippocampal volume (right+left) was smaller in PTSD subjects as well (9.4%), but the difference was also not significant (8.326±1.073 cm³ vs. 9.11±1.085 cm³, \(P>0.05\)).
PTSD subjects had smaller right than left hippocampus (7.9%), and the difference was statistically significant (4.017±0.527 cm³ vs. 4.33±0.541 cm³; t=8.250, P=0.000). Normal control subjects had smaller left than right hippocampus (5.61%), and the difference was statistically significant (4.428±0.521 cm³ vs. 4.682±0.587 cm³; t=−8.124, P=0.000).

4. Discussion

In the current study, we compared hippocampal volumes in 15 right-handed patients with chronic combat-related PTSD and case-matched healthy subjects. Our results show significantly smaller right hippocampus in subjects with PTSD (13.5%). Also, the left hippocampus was smaller in PTSD subjects (1.1%), but the difference was not statistically significant. Further on, in all PTSD subjects, the right hippocampus was smaller than the left (on average, 7.88%). In all normal control subjects, the right hippocampus was bigger than the left (5.61%). Our results suggest decreased right hippocampal volume in our PTSD subjects.

Some earlier studies found lower bilateral hippocampal volumes in patients with PTSD than those observed in control groups (Gurvits et al., 1996; Villarreal et al., 2002; Bremner et al., 2003; Hedges et al., 2003). Others have found hemispheric asymmetries in such differences, with significantly greater reduction in either the right (Bremner et al., 1995; Shin et al., 2004; Wignall et al., 2004; Winter and Irle, 2004) or the left (Bremner et al., 1997; Stein et al., 1997; Nakano et al., 2002; Lindauer et al., 2004) hemisphere. Other studies have failed to find any such differences (De Bellis et al., 1999, 2002; Bonne et al., 2001; Carrion et al., 2001; Schuff et al., 2001; Fennema-Notestine et al., 2002; Pederson et al., 2004).

Such diversity in results is probably a consequence of highly heterogeneous groups that have been spanned in these studies, regularly with small sample sizes (Kitayama et al., 2005; Smith, 2005). Studies are performed either on children or adults, focusing primarily or exclusively on a single gender vs. studies including both genders, patients suffering from stress associated with a wide variety of different traumatic experiences (e.g., combat exposure, childhood physical or sexual abuse, and medical trauma), differences in comorbid conditions, differences in the nature of the control groups used as a basis of comparison. A significant source of uncertainty has also been contributed by variations in imaging methods and volumetric analysis procedures between laboratories (Pedraza et al., 2004; Smith, 2005). Between-study variability in hippocampal volume estimates has been shown to be much greater than the differences in hippocampal volumes between the compared subject groups within individual studies.

Our results are at least partially consistent with a recent neuroanatomical model of PTSD in which an abnormally functioning hippocampus and medial prefrontal cortex fail to inhibit a hyperresponsive amygdala (McNaughton, 1997; Rauch et al., 2000; Shin et al., 2001; Hull, 2002). There are several possible explanations for smaller hippocampal volume in PTSD. Stress is associated with several factors that could be implicated in hippocampal neuronal damage and subsequent reduction in volume, including increased glucocorticoid secretion, inhibition of neurogenesis, and reductions in brain-derived neurotrophic factor (Kitayama et al., 2005; Smith, 2005). Still the major role is attributed to excessive glucocorticoid secretion, but the evidence for that is inconsistent at best. Patients with PTSD consequent to combat stress have sometimes been found to have relatively low levels of urinary cortisol in comparison with control subjects, possibly reflecting dysfunction in adrenal–cortical sensitivity to feedback regulation (Yehuda et al., 1991, 1995; Yehuda, 2001).

Further on, it is also possible that subjects with smaller hippocampal volume are predisposed to develop PTSD following exposure to a traumatic event (Gilbertson et al, 2002). It is known that a considerable portion of the variance in PTSD is genetic (Goldberg et al., 1990).

Two recent meta-analyses of studies that used volumetric analyses of structural MRI data to examine hippocampal volumes in patients with PTSD symptoms both found bilateral volume reduction relative to those in normal control subjects and traumatized controls with no PTSD (Kitayama et al., 2005; Smith, 2005). In the present study, we found a significant reduction only on the right side. While the left hippocampi were also smaller in PTSD subjects, the difference was not found statistically significant. Although some other studies have also found a significant hippocampal reduction only in the right hemisphere, our finding may well be a result of lack of the experimental power in our study and might change with larger subject samples (Bremner et al., 1995; Shin et al., 2004; Wignall et al., 2004; Winter and Irle, 2004). On the other hand, results from a recent meta-analysis of the normal population parameters of hippocampal and amygdala asymmetry and absolute intrahemispheric volumes strongly suggest that the hippocampi are asymmetrical structures with larger volumes in the right hemisphere (Pedraza et al., 2004). They report mean right hippocampal volume of 3.06 cm³ (S.D. = 1.01 cm³) and mean left hippocampal volume of 2.98 cm³ (S.D. = 0.98 cm³), measurements
that are compatible with our findings in the normal control group. If and how this natural right-to-left asymmetry in hippocampal volumes is associated with the asymmetry in hippocampal volume reduction in our results is impossible to say at this moment.

The strength of present study is in the selection of the participants. We selected a very homogeneous PTSD population, regarding the type and time of traumatic experience, and the time period between traumatization and inclusion in the study. Earlier studies have shown that these factors can influence the clinical presentation and possibly also the etiology of PTSD (Henigsberg et al., 2001; Gregurek et al., 2001; Kozaric-Kovacic et al., 2004). We believed that long enough period following the traumatization should allow putative patho-physiological changes in hippocampal structures to advance sufficiently to produce morphological changes that could be measured using our methodology. Note that our subjects received no psychiatric or psychological treatment and no psychotropic medication during this period, factors that have been shown to influence hippocampal volume (Sheline et al., 2003; Vermetten et al., 2003). Second, we excluded comorbid conditions that could have influenced the findings, especially depression and substance abuse (Frodal et al., 2002; Laakso et al., 2000). Furthermore, all our PTSD subjects were normally functioning in society before the traumatization. Also, our study was a case-matched control study with healthy subjects as controls and with age, gender, handedness, years of education, and socioeconomic level as matching factors.

The basic limitation of the current study is that we had to use normal controls rather than combat controls with similar traumatic experiences and no PTSD. Use of a control group with combat exposure would have allowed us to dissociate the traumatic experience per se from the development of PTSD, as possibly associated with reduced hippocampal volume. Thus, an important question remains unanswered, but it was technically impossible for us to find such control subjects primarily because of a very small number of possible subjects all together. Croatia is a small country with a total population of around 4.5 million and consequently small armed forces. Moreover, to identify other soldiers with similar experiences, we would have to have had access to the military records, which we did not for legal reasons. On the other hand, we wanted our subject population to be as homogeneous as possible for the reasons described above.

Despite any shortcomings, we believe our results strongly indicate decreased hippocampal volumes in subjects with chronic combat-related PTSD, at least in the right hemisphere. Some important questions still remain unanswered. First, does reduced hippocampal volume constitute a predisposition to or a consequence of the development of PTSD following traumatic psychological experience? Second, is the normal right-to-left asymmetry in hippocampal volume associated with right-to-left asymmetries in hippocampal volume reduction in some studies (including ours), and if so in what way? Final resolution of these issues will depend on future longitudinal studies that sample hippocampal volume both before and after exposure to different types of stressors and putative development of PTSD.

References


