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Neuroendocrine and immune responses to a cognitive stress challenge in veterans with and without PTSD

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Background: PTSD has been associated with altered hypothalamus–pituitary–adrenal-axis (HPA-axis), immune and sympathetic nervous system (SNS) regulation. The purpose of this study was to evaluate the effect of cognitive stress on these systems in PTSD patients and controls.

Methods: The subjective units of distress score (SUDS), NK-cell response, plasma levels of noradrenalin and ACTH in response to cognitive stress were assessed in male veterans with PTSD ($n = 15$) and age, region and year of deployment matched veterans without psychopathology ($n = 15$).

Results: The challenge induced an increase in SUDS, noradrenalin, ACTH and NK-cell response in both groups. Baseline levels of ACTH were lower in PTSD patients. The test was experienced as more stressful by PTSD patients and resulted in an augmented ACTH response in patients. The noradrenalin and NK-cell responses showed no group differences. The ACTH response correlated with the severity of symptoms in patients, and the noradrenalin response correlated with the ACTH and NK-cell response in controls, but not in patients.

Discussion: PTSD patients experience more distress and present with an exaggerated pituitary response to this stressor. In addition, our results suggest an altered interaction between the HPA-axis, SNS and immune system in PTSD.

Keywords: *cognitive stress challenge*; *PTSD*; *HPA axis*; *NK-cell activity*; *catecholamines*; *ACTH*; *cortisol*

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Functional interactions between the sympathetic nervous system (SNS) and the hypothalamic–pituitary adrenal axis (HPA-axis) are required for appropriate responses to stress, anxiety and fear (Vermetten & Bremner, 2002). Both systems have been extensively studied in posttraumatic stress disorder (PTSD) and the data provide evidence for a dysregulation of both the SNS and HPA axis in PTSD. Thus, studies measuring baseline levels of noradrenalin (NA) in the cerebrospinal fluid (CSF) have reported elevated levels that correlated with PTSD symptoms (Geracioti et al., 2001; Strawn, Ekhator, Horn, Baker, & Geracioti, 2004). Assessments of baseline HPA-axis function in PTSD have shown elevated levels of CRF in CSF (Baker et al., 1999; Hovens, Bramsen, & Van der Ploeg, 2002; Sautter et al., 2003) and plasma (De Kloet et al., 2008). To assess the reactivity of both systems to psychological stress, several challenge paradigms have been employed. The most commonly used test is the Trier Social Stress Test (TSST) (Kirschbaum, Pirke, & Hellhammer, 1993).

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Stressful challenges that also have been utilized in PTSD, include trauma scripts (Elzinga, Schmahl, Vermetten, Van Dyck, & Bremner, 2003), traumatic reminders (Blanchard, Kolb, Prins, Gates, & McCoy, 1991; Liberzon, Abelson, Flagel, Raz, & Young, 1999; McFall, Murburg, Ko, & Veith, 1990) and mental arithmetic tasks (Bremner et al., 2003; Vermetten et al., 2006). Studies in PTSD have shown increased SNS responses to trauma related stimuli (Blanchard et al., 1991; Liberzon et al., 1999; McFall et al., 1990), and elevated salivary cortisol levels in anticipation, but not in response to traumatic scripts (Valentino, 1988), traumatic reminders (Liberzon et al., 1999) and arithmetic tasks (Dunn, Swiergiel, & Palamarchouk, 2004). No differences in cortisol were reported in response to the TSST in patients with PTSD compared to healthy controls (Roelofs et al., 2009; Simeon, Knutelska, Yehuda, Putnam, & Schmeidler, 2007).

Animal research has provided evidence for mutual interactions between the HPA-axis and the SNS. The major NA containing nucleus in the brain, the Locus Coeruleus (LC), receives CRH afferents, and central administration of CRH into the LC has shown to increase the LC firing and NA release in its projection areas (Jedema & Grace, 2004; Valentino, 1988). LC neurons also provide noradrenergic input to the paraventricular nucleus of the hypothalamus leading to HPA-axis activation (for review, see Dunn et al., 2004). Studies in humans on the relationship between HPA-axis and SNS responses to stress, however, are scanty (Young, Abelson, & Cameron, 2005).

There is increasing evidence that psychological stress also influences the susceptibility to inflammatory and infectious diseases (Black & Garbutt, 2002; Jessop, Richards, & Harbuz, 2004; Marshall, 2004). Natural Killer (NK) cell activity, relevant for protection against pathogens and malignant transformation (for review, see O'Connor, Hart, & Gardiner, 2006), have shown to increase following psychological stressors (Benschop, Rodriguez-Feuerhahn, & Schedlowski, 1996; Benschop et al., 1998; Segerstrom & Miller, 2004). This effect is known to be mediated by the SNS (for review, see Sanders & Straub, 2002). This relation between NK-cell activity and the SNS is strengthened by reports on the relationship between NK-cell activation on the one hand and the catecholamine secretion and heart rate and blood pressure increases in response to stress on the other (for review, see Sanders & Straub, 2002).

To evaluate the effect of cognitive stress on the SNS, HPA-axis and immune response in PTSD, we measured plasma NA, ACTH levels and the NK-cell activity in veterans with PTSD and equally trauma exposed veterans without PTSD.

Epidemiological surveys indicate that the vast majority of individuals with PTSD also meet criteria for at least one other psychiatric disorder (Brady, Killeen, Brewerton, & Lucerini, 2000; Brunello et al., 2001). The most common co-morbid diagnosis is major depressive disorder (MDD). In view of the high incidence of co-morbid MDD in PTSD (Shalev, 2001), and the substantial overlap in clinical symptoms between the two conditions, we decided not to exclude patients with a co-morbid MDD.

Methods

Participants

PTSD patients were recruited from the Department of Military Psychiatry at the Central Military Hospital, Utrecht, The Netherlands. All new patients diagnosed with PTSD between August 2002 and August 2004, were invited to participate in this study. Controls were selected from a group of male veterans registered at the "Veterans Institute (VI)". They were matched with the PTSD group on age, year and region of deployment. All veterans were screened for psychiatric illness using the Structured Clinical Interview for DSM IV axis I disorders (SCID-I, First, Spitzer, Gibbon, & Williams, 1997). The diagnosis of PTSD was confirmed by the Clinical Administered PTSD Scale (CAPS) (Blake et al., 1995) and after consensus by three clinicians (C.d.K, A.R, E.G). Only PTSD patients with a CAPS score above 50 were included. PTSD patients with a psychiatric disorder other than mood and anxiety disorders were excluded. Controls were included if they met the A1 criterion for PTSD, but had a CAPS score below 25 and did not meet DSM IV criteria for PTSD or any other current DSM IV axis I disorder. Severity of depressive symptoms and anxiety were assessed using the Hamilton Depression Rating Scale (HDRS) and the Hamilton Anxiety Scale (HAS). None of the included veterans were physically injured during their deployment.

Subjects who had a history of neurological illness or a significant medical illness were excluded. The study was approved by the Institutional Review Board of the University Medical Center Utrecht, The Netherlands. Written informed consent was obtained from all subjects who participated in the study after a complete written and verbal description of the study.

Challenge procedure

The design of the challenge test was adopted from other studies in PTSD patients (Dunn et al., 2004; Vermetten et al., 2006). In summary, the challenge protocol consisted of a relaxation phase followed by a phase in which the participant was invited to perform a serie of cognitive tasks (including memory tests for figure number pairing, a problem solving tasks and an arithmetic test (subtraction) under time pressure. All subjects were studied in the time interval from 13.00 to 15.30 h.

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The challenge phase was followed by a relaxation phase and debriefing.

The participants arrived at the research unit at 12.30 h. They were asked to rest supine on a bed. A cannula was inserted in the forearm vein and kept open with NaCl. Subjects rested for 1 h while listening to relaxing music. After baseline sampling (t1) at 13.55 h, for measurement of plasma ACTH, NA levels and percentage of NK lysis (NK-cell activity), they were invited to sit at a table waiting for the investigator to arrive. At this time the subjective units of distress scale (SUDS) was administered. This scale measured the amount of distress on a scale ranging from 0 to 100 (0 = no stress, 100 = maximal stressed). At the start of the challenge an investigator that was unknown to the subject entered the room, went behind the desk, and initiated the tasks mentioned above. The tests lasted for 20 min and were completed at 14.20 h. Negative feedback regarding the score and the time spent in the task, was consistently given, and the level of difficulty was increased until subjects were unable to successfully complete the tasks.

During the challenge, blood was sampled for measurement of ACTH and NA at 14.05 h (t2), 14.10 h (t3), 14.15 h (t4), 14.20 h (t5), and for NK-cell activity at 14.20 h. At the end of the test the investigator left the room and patients rested in supine position for another hour on a bed. They were debriefed about the design of the test. Directly after the test, the SUDS was administered for the second time and patients were asked to rest on the hospital bed for another hour with relaxing music. At 15.20 h (t6) blood was sampled, for measurement of ACTH, NA and NK-cell activity. After this last blood sample the cannula was removed and the SUDS was administered for the third time.

Biochemical analysis

Blood samples for the measurement of ACTH and NA measurement were collected in EDTA tubes, immediately put on ice and centrifuged at 4°C. Blood plasma was separated and immediately stored at −80°C until analysis. The determination of adrenocorticotropic hormone (ACTH) was performed with a solid-phase, two-site sequential chemiluminescent immunometric assay (Immulite 2500, DPC, Los Angeles, USA) with a detection limit of 5 ng/l. NA were measured using high performance liquid chromatography (HPLC) with electrochemical detection (ClinRep).

Heparinized venous blood was collected for measurement of NK-cell function. NK-cell activity was determined in a standard ^{51}Cr release assay with K562 cells as target cells. Whole blood was incubated with 10,000 ^{51}Cr-labeled K562 cells for 4 h. Total and background release of ^{51}Cr was determined by adding100 µl 1% Triton-X100 or medium, respectively. NK-cell activity was calculated as: % killing = ((cpm sample cpm background)/(cpm total cpm background)) × 100.

Statistical analysis

To measure the response to the challenge paradigm a derivative parameter, the area under the plasma concentration time curve with respect to response (AUCr), was calculated using the trapezoidal formula previously described by Pruessner, Kirschbaum, Meinlschmid, and Hellhammer (2003). Subjective distress of the test was measured by calculating the increase in SUDS between t1 and t5 (delta SUDS).

To properly analyze the effects of all variables on the responses of interest, multivariate analysis (MANOVA), with baseline levels (t1) of ACTH, NA and NK lysis, the AUCr of the plasma ACTH, NA and NK lysis levels between t1 and t5 and delta SUDS as dependent variables and group as fixed factor, was used to look for significant group differences in baseline levels and response to challenge. Whenever necessary, data were log-transformed to normalize the distribution. In addition the responses to the challenge paradigm and the recovery after the test were analyzed using analysis of variance with repeated measures on time.

Within groups correlation analyses (Spearmann correlation) were performed between CAPS total score, SUDS scores, and the AUCr of all outcome measures.

In addition, the effects of a current co-morbid MDD were analyzed with MANOVA comparing all outcome measures between three groups (PTSD patients with and without MDD and healthy controls).

All statistical analyses were performed with SPSS 12.0 for Windows (SPSS, Chicago, IL). Statistical threshold for significance for all measures was set at $p < 0.05$.

Results

Demographics

The demographic characteristics of all PTSD patients and controls are displayed in Table 1. Patients and controls did not differ in body mass index or race. Thirteen out of 15 patients were naïve for psychotropic medication and all other patients were medication free for at least 4 weeks. Patients with PTSD had significantly higher CAPS, HAS, and HDRS scores, (see Table 1). The mean CAPS score in PTSD patients was 73.7 (range 55–89) and in controls 7.4 (range 0–18). Patients and controls were exposed to similar traumatic events (Table 2). Lifetime (past) DSM-IV diagnosis of the PTSD patients and controls are displayed in Table 1. Seven subjects with PTSD also met current diagnostic criteria for MDD, one subject met current diagnostic criteria for somatoform disorder and one subject with PTSD met current diagnostic criteria for panic disorder

Table 1. Demographics characteristics and test variables of PTSD patients and controls. Variables displayed as mean standard deviation and range

	PTSD ($n=15$)			Controls ($n=15$)			
	Mean	SD	Range	Mean	SD	Range	*P*
Age (years)	34.9	6	27–44	34.0	5.1	26–33	$p=0.78$
Race	Caucasian	$n=15$		Caucasian	$n=15$		
Smoking >3 sig/day	$n=8$			$n=4$			
Year deployment	1990	5.9	1981–1995	1991	5.7	1980–1996	$p=0.84$
Country deployment	Bosnia $n=9$			Bosnia $n=10$			
	Lebanon $n=5$			Lebanon $n=5$			
	Cambodia $n=1$						
CAPS	73.7	10.9	55–89	7.4	6.7	0–18	$p<0.001$*
HDRS	15	4.7	9–22	1.5	1.6	0–5	$p<0.001$*
HAS	17.4	6.3	5–28	1.9	1.6	0–5	$p<0.001$*
Comorbid disorders (lifetime)	MDD ($n=9$)			MDD ($n=3$)			
	Bipolar II disorder ($n=3$)			Panic disorder ($n=1$)			
	Alcohol dependence ($n=1$)						
	Alcohol abuse ($n=2$)						
	Substance abuse ($n=1$)						
	Panic disorder ($n=1$)						
	Somatoform disorder ($n=1$)						

*Significant differences between groups.

Table 2. Traumatic events during deployment. The table displays the number of PTSD patients and trauma controls that were exposed to, or eyewitness of, these events, as assessed with the CAPS

	PTSD ($n=15$)		Trauma controls ($n=15$)	
Event	Experienced	Witnessed	Experienced	Witnessed
Fire or explosion	9	2	9	1
Transportation accident	1	3	4	1
Physical assault	8	3	4	0
Assault with a weapon	13		14	
Sexual assault		1		
Other unwanted sexual experience		1		
Combat or exposure to a war-zone	14		14	
Captivity (held hostage)	6		4	
Life threatening illness or injury	1	8		5
Severe human suffering		14		13
Witnessing sudden violent death	11		4	
Sudden, unexpected death of someone close to you	5		6	
Serious injury or death you caused to someone else	4		1	
Witnessing dead bodies, human remains and/or massgraves	5		8	

with agoraphobia. The SCID did not reveal any current psychiatric disorders in the control group.

Cognitive stress challenge

MANOVA showed significant group differences ($F=3.5$, $p=0.012$). With significant differences in increase of SUDS (delta SUDS), baseline ACTH levels and AUCr of plasma ACTH concentrations during the challenge. No significant differences in baseline or AUCr of plasma NA levels and NK cell-lysis were observed.

Figure 1A displays the time-course of the subjective units of distress scores (SUDS) for patients and controls during and after the test. ANOVA showed no significant group difference in SUDS before the challenge (Table 3). The challenge resulted in a significant increase in SUDS in both group ($F=75$; $p<0.001$). Statistical analysis showed a significant group by time interaction with a greater increase in SUDS in patients compared to controls ($F=8.3$, $p=0.008$). The SUDS returned to baseline level within an hour after the test in both groups. ANOVA showed no group differences in SUDS at end point (Table 3).

The time-courses of plasma ACTH levels before, during and after the challenge is displayed in Fig. 1B, ANOVA showed that at baseline ACTH levels were significantly higher in PTSD patients compared to controls ($p=0.02$) (Table 3). The challenge induced a statistically significant increase in plasma ACTH ($F=29$; $p<0.001$) levels in both groups. Analysis of variance with repeated measure on time showed a significant group by time interaction between t1 and t5 ($F=4.3$; $p=0.038$). Delta SUDS was not a significant covariate. ANOVA showed that the AUCr of plasma ACTH levels was significantly higher ($p=0.007$) in patients compared to controls (Table 3). No significant group by time interaction was observed in ACTH levels in the recovery phase (t5–t6). ANOVA showed that at endpoint the plasma ACTH ($p=0.04$) levels were still significantly higher in PTSD than in controls (Table 3).

In Fig. 1C,D the time-courses of plasma NA levels and NK-cell activity during and after the challenge are displayed. ANOVA showed that baseline NA levels as well as NK-cell activity did not show significant differences between groups (Table 3). The challenge induced a statistically significant increase in plasma NA ($F=3.3$; $p=0.03$) and NK-cell activity ($F=79$, $p<0.001$). Analysis of variance with repeated measure on time showed no significant group by time interaction during the challenge (between t1 and t5) and in the recovery phase (t5–t6) in plasma NA and NK-cell activity. ANOVA showed that at endpoint no significant differences were observed in plasma NA levels and NK-cell activity either (Table 3).

In PTSD patients the CAPS total score correlated significantly with the ACTH response (AUCr) ($r=0.60$, $p=0.02$) (Table 4). The increase in SUDS also correlated significantly with NK-cell function in patients ($r=0.55$, $p=0.04$), but not in controls ($r=-0.38$, $p=0.17$) (Table 4). No correlations were observed between all other outcome measures and the SUDS, neither in patients nor in controls. The plasma NA response correlated significantly with the ACTH ($r=0.53$; $p=0.05$) and NK-cell ($r=0.61$; $p=0.02$) response in controls, but not in patients (ACTH: $r=-0.10$; $p=0.71$; NK-cell: $r=0.01$; $p=0.95$) (Table 4).

When patients were subdivided into subjects with and without a current co-morbid MDD, MANOVA

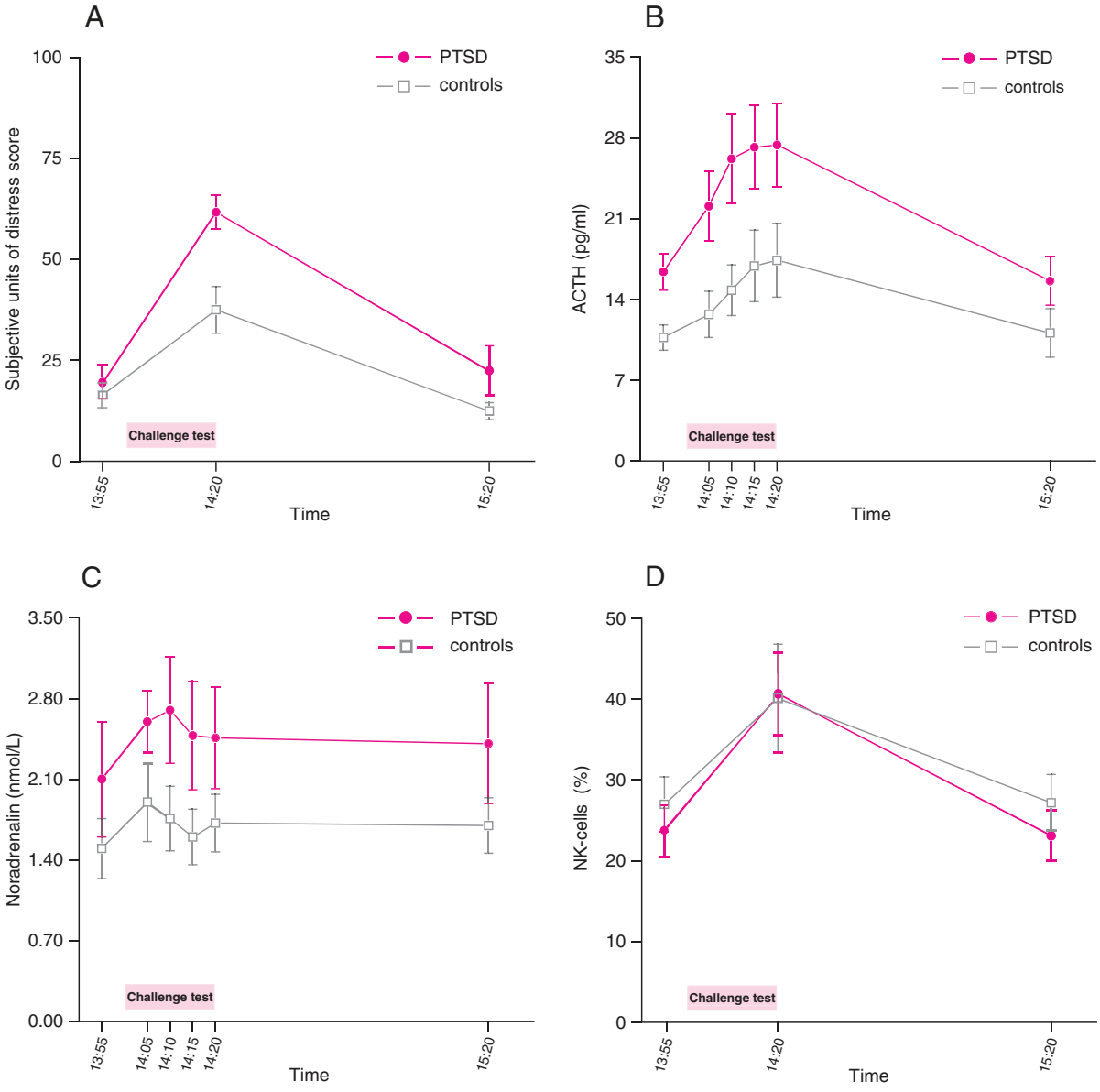

Fig. 1. Subjective Units of Distress score (1A), ACTH (1B), noradrenalin (1C) levels and NK-lysis (1D) before, during and after the cognitive stress challenge in PTSD patients and controls.

showed no significant differences between patients with and without a co-morbid MDD on all outcome variables.

Discussion

Psychological stress as used in this study induced feelings of distress in all subjects, but the effect was more pronounced in patients with PTSD than in trauma controls. This cognitive stress challenge also induced significant increases in plasma NA levels and in ACTH and NK-cell response in all subjects, but only the ACTH response was significantly larger in patients as compared to controls, suggesting an enhanced sensitivity of the HPA-axis to psychological stress in PTSD.

In previous studies salivary or plasma cortisol levels have been assessed to measure the effect of psychological stress on the HPA axis. In three out of six studies elevated cortisol levels were reported in anticipation to but not in response to the challenge in PTSD patients compared to healthy controls (Bremner et al., 2003; Elzinga et al., 2003; Liberzon et al., 1999). Only one study also reported elevated cortisol levels in response to the challenge (Vermetten et al., 2006). These studies already suggested that despite the frequently reported low baseline levels of cortisol, the cortisol sensitivity to, or in anticipation of, a psychological stressor is not impaired in PTSD. The present study shows significantly higher baseline plasma ACTH levels and higher ACTH levels in response to

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Table 3. Subjective distress, noradrenalin, ACTH and Natural Killer (NK) cell responses to the cognitive stress challenge

	PTSD			Trauma controls			
	Median	Mean	SD	Median	Mean	SD	Subject group effect
SUDS							
t0	15.0	19.40	15.80	17.5	16.30	11.70	$F(1,26) = 0.50$, $p = 0.49$
t5	60.0	61.70	15.80	40.0	37.50	21.80	$F(1,27) = 11.79$, $p = 0.002$*
t6	15.0	22.4	23.8	12.5	12.4	8.00	$F(1,27) = 2.25$, $p = 0.15$
ACTH (pg/ml)							
t0	15.4	16.4	6.2	10.6	10.70	4.30	$F(1,26) = 6.70$, $p = 0.02$*
AUCr t0–t5	74.0	159.60	192.6	27.0	40.50	61.60	$F(1,26) = 8.63$, $p = 0.007$*
t6	13.5	15.60	8.00	8.6	11.10	8.00	$F(1,28) = 4.67$, $p = 0.04$*
Noradrenalin (nmol/l)							
t0	1.6	2.1	1.30	1.2	1.48	1.01	$F(1,26) = 2.31$, $p = 0.14$
AUCr t0–t5	4.7	8.2	18.8	1.6	4.9	12.4	$F(1,26) = 0.10$, $p = 0.76$
t6	1.8	2.4	0.89	1.7	1.74	1.97	$F(1,26) = 1.07$, $p = 0.31$
NK cell lysis (%)							
t0	22	23.7	12.5	25	27	13.3	$F(1,26) = 0.05$, $p = 0.83$
AUCr t0–t5	140	169.00	108.00	120	131	204	$F(1,26) = 0.01$, $p = 0.95$
t6	22	23	12	24	27	13	$F(1,27) = 0.77$, $p = 0.39$

Note: Baseline levels (t0), levels after recovery (t6) and area under the response curve (t0–t5) were whenever necessary log-transformed and compared using ANOVA. Variables are displayed as median, and mean.
*Significant differences between groups.

stress in PTSD patients. ACTH response correlated significant with CAPS scores in patients. Patients experienced the challenge as more stressful than controls as manifested by a higher SUDS score. However, no correlations were observed between the SUDS score on the one hand and ACTH responses on the other, neither in patients nor in controls, suggesting that the psychological and neuroendocrine responses to the psychological stress are unrelated. This would imply that the exaggerated HPA-axis response in patients is not accounted for by differences in subjective experienced distress but suggests an enhanced HPA-axis sensitivity in PTSD to psychological stress. In accordance with this explanation, Vermetten et al. (2006) found an augmented salivary cortisol response to a cognitive challenge in PTSD patients before—as compared to the response after—long-term treatment with paroxetine, despite similar SUDS scores. In the only other study that also assessed the ACTH response, no differences were observed in ACTH levels in anticipation to and in response to traumatic reminders (Liberzon et al., 1999). The fact that ACTH levels remained significantly higher in PTSD patients until at least one hour after the challenge procedure is of specific interest and suggests a prolonged HPA-axis activity in PTSD. This phenomenon was, however, not reported in other cognitive stress challenges.

In previous studies elevated baseline levels of catecholamines (Liberzon et al., 1999; Yatham, Sacamano, & Kusumakar, 1996) and an exaggerated NA response to

Table 4. Correlations (Spearmann's row) between PTSD symptoms, noradrenalin, ACTH and NK cell responses (AUCr) in PTSD patients and controls

	Group	CAPS	AUCr ACTH	AUCr NA
AUCr ACTH	TC	$r = .29$; $p = 0.31$		
	PTSD	$r = 0.60$; $p = 0.02$*		
AUCr NA	TC	$r = 0.26$; $p = 0.47$	$r = 0.53$; $p = 0.05$*	
	PTSD	$r = -0.08$; $p = 0.77$	$r = -0.10$; $p = 0.71$	
AUCr NKlysis	TC	$r = 0.23$; $p = 0.40$	$r = 0.56$; $p = 0.04$*	$r = 0.61$; $p = 0.02$*
	PTSD	$r = 0.20$; $p = 0.47$	$r = 0.39$; $p = 0.15$	$r = 0.01$; $p = 0.99$

*Significant differences between groups.

traumatic reminders; (Liberzon et al., 1999; McFall et al., 1990; Yatham et al., 1996) have been reported in PTSD. This finding could not be replicated in the present study using a cognitive challenge test. Differences in design and a lack of statistical power in the present study might explain this discrepancy.

Animal studies have provided evidence for a relationship between the HPA-axis and SNS (for review, see Carrasco & Van de Kar, 2003; Young et al., 2005). A limited number of studies have reported on the relationship between the HPA-axis and the noradrenergic system in humans (O'Donnell, 2004). The majority of these studies reported a greater HPA-axis or NA activation following stress, suggesting a relationship between HPA-axis and SNS reactivity. In line with this notion, we found a significant correlation between the NA and ACTH response in control subjects. In patients with PTSD, however, this relationship seems to be vanished. This is in accordance with the only other study in PTSD that has assessed the catecholaminergic and HPA-axis response to a psychological stressor (Liberzon et al., 1999). In this study no significant correlations were found between the HPA-axis and catecholamine responses as well.

In the present study no significant group differences were observed in baseline and stressor-induced NK-cell function. NA is known to regulate NK-cell function. Studies in healthy subjects have shown high correlations between NK-cell function and NA responses to stress. Indeed, in our control group the NA response to the stressor correlated significantly with NK-cell response, suggesting that trauma exposure *per se* had not altered the relationship between catecholamine and NK-cell activity. In patients, however, this relationship was no longer present suggesting an altered regulation of NK-cell activity by catecholamines in PTSD.

This study has some limiting factors. First of all, interpretation of peripheral ACTH and NA concentrations and dynamics should be cautious, as peripheral levels may not well reflect those in CNS. Second, the number of included subjects is low. The inclusion was limited by stressfulness of the procedure. Third, smoking habits, which may affect cortisol levels (Olff et al., 2006), were not taken into account. A limitation of the challenge design is that the amount of social support could be influenced by the reaction of the participant.

In conclusion, we found higher ACTH levels in the anticipation phase and an exaggerated ACTH response to a cognitive challenge in PTSD patients compared to trauma controls. No differences in reactivity to psychological stress were observed in NA response and NK-cell reactivity. No relationship was found between the neuroendocrine responses and amount of distress experienced by the subjects. To evaluate the role of trauma exposure, future studies should include a non-traumatized control group. In addition it would be of interest to assess NA, cortisol and ACTH levels in cerebrospinal fluid in response to a cognitive stressor.

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Conflict of interest and funding

There is no conflict of interest in the present study for any of the authors.

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