Sleep disturbances and post-traumatic stress disorder; A perpetual circle?

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Sleep disturbances and post-traumatic stress disorder; A perpetual circle?

Slaapstoornissen en posttraumatische stressstoornis: een vicieuze cirkel?

(met een samenvatting in het Nederlands)

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Chapter 1

Introduction and general outline

"Sleep is like food and medicine for the sick"
Pirkei de Rabbi Eliezer

This dissertation focuses on sleep in patients with a combat-related post-traumatic stress disorder (PTSD). In this chapter, we will shortly introduce PTSD, sleep, sleep-dependent synaptic plasticity, memory consolidation, and the possible bidirectional relationship between sleep and PTSD. Subsequently, we will formulate our research questions and the general outline of this dissertation.

SLEEP-RELATED BRAIN ACTIVITY

Recent research has shown that sleep is not merely a resting phase in which brain activity shuts down. On the contrary, during Rapid Eye Movement (REM) sleep the brain is as active as during waking, and uses as much energy as in the waking state (Buchsbaum et al., 2001). Other brain activity patterns are observed during REM sleep, with high activity in limbic structures and low activity in prefrontal areas (Maquet et al., 2005). During non rapid eye movement (NREM) sleep, reactivation of the hippocampus has been demonstrated (Wilson and McNaughton, 1994), a brain area in which neurogenesis continues to take place during adult life (Eriksson et al., 1998). Moreover, reactivation of neurons in the hippocampus during slow wave sleep enhances memory retention (Rasch et al., 2007b; Peigneux et al., 2004). A wealth of literature shows the importance of sleep on learning in humans, suggesting neural plasticity during sleep (Wang et al., 2011). Animal studies have shown that sleep regulates neurogenesis and genes associated with neural plasticity (Cirelli and Tononi, 2000; Mueller et al., 2011; Rasch et al., 2007a). Sleep ensures us to wake up with a 'slightly different brain' than the night before. Sleep-depended neuronal plasticity may be of importance in psychiatric disorders. Firstly, because psychiatric disorders have been associated with a decrease in neurotrophic factors (Lewis and Gonzalez-Burgos, 2008; Manji et al., 2003). Secondly, because disturbed sleep in psychiatric disorders may compromise treatment-effect (Troxel et al., 2011), possibly by hampering synaptic plasticity during sleep. Reduced synaptic plasticity during sleep may possibly act as a precipitating or a perpetuating factor in psychiatric disease.

In most psychiatric disorders, decreased hippocampal volumes have been demonstrated (Geuze et al., 2005). Interestingly, insomnia severity is inversely related to hippocampal volume in patients with a posttraumatic stress disorder (Neylan et al., 2010), and in patients suffering from primary insomnia (Riemann et al., 2007). Reduced hippocampal activity during a memory task was also observed as a result of sleep deprivation (Yoo et al., 2007) and sleep fragmentation (Van Der Werf et al., 2009).

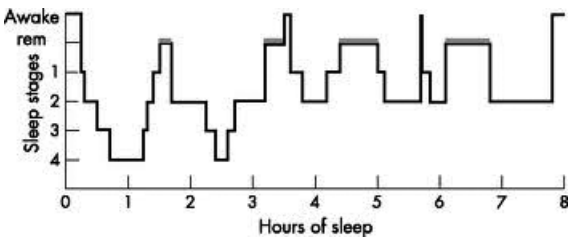
The evidence of a relationship between plasticity and sleep on the one hand, and plasticity and psychiatric disorders on the other hand is emerging. So far, in psychiatric research the neurobiology and consequences of disturbed sleep on plasticity, learning and recovery are relatively rarely studied phenomena.

SLEEP AND POST-TRAUMATIC STRESS DISORDER

Post-traumatic stress disorder (PTSD) is related to a high prevalence of insomnia and nightmares (Neylan et al., 1998; Ohayon and Shapiro, 2000). It is beyond doubt that peaceful sleep is affected in PTSD patients. However, cross-sectional polysomnographic (PSG) studies on objective sleep quality in PTSD leave the researchers in the field puzzled, since these showed only mildly disturbed objective sleep quality (Breslau et al., 2004; Hurwitz et al., 1998; Kobayashi et al., 2007; Pillar et al., 2000). A discrepancy was seen between the severity of the subjective complaints reported by patients and the relatively healthy amount of objective total sleep time (TST). (For an overview of normal sleep architecture see box 1).

In order to understand how sleep may be affected in PTSD it is important to know a little more about the phenomenology of PTSD and the neurobiological changes associated

BOX 1: Sleep architecture.



Sleep consists of rapid eye movement sleep (REMS), and Non REMS (NREM). NREMS consists of stage (S)1, S2, S3 and S4 sleep. See the figure in this box. The different sleep stages can be determined with *polysomnography* which consists of simultaneous recordings of the electro-encephalogram (EEG), electro-oculogram (EOG), electro-myogram (EMG) of the chin muscle, and often an electro-cardiogram (ECG). EEG measures brain activity, which differs between sleep stages. S1 is the lightest phase of sleep. It is often a short period between wake en S2 sleep. S2 sleep comprises about 50% of our total sleep period, it also referred to a "light NREM" sleep. Delta activity on the EEG (slow waves between 0.5 and 4 Hz) predominate S3 and S4. Therefore, S3 and S4 are often referred to as slow wave sleep (SWS) or delta sleep*. The EEG activity during REM sleep may resemble wake. Therefore, muscle paralysis as determined on the EMG and rapid eye movements on the EOG are measured to distinguish REM sleep from wake. SWS occur in the first half of the night. During SWS brain metabolism is at its lowest. A normal night sleep comprises of approximately 50% NREM2, 25% REM, 20 % SWS, and 5% S1 sleep.

* According to new guidelines of the American Academy of Sleep Medicine (AASM), S3 and S4 sleep are referred to as N3. However, in this manuscript sleep was scored according to criteria by Rechtschaffen and Kales (1968).

with this disorder. PTSD is an anxiety disorder which is directly related to a traumatic experience. It is characterized by frequent re-experiencing of the traumatic event, irritability, anxiety symptoms and avoidance of stimuli that trigger fear or unpleasant emotional reactions. PTSD can have a chronic and therapy-resistant course (Johnson et al., 2004; Solomon and Mikulincer, 2006). In the Netherlands, the lifetime prevalence of PTSD is 7-8 % (de Vries and Olff, 2009).Those who are professionally exposed to violence are at risk of developing PTSD.

While the prevalence of PTSD may not be as common as depression (20% life time prevalence) in civilian populations (Kessler et al., 1994), PTSD is a frequent and relevant disorder in military psychiatry. In the Netherlands, about 100.000 military personnel have participated in a range of military deployments since the Second World War. Amongst the most well-known are Lebanon (1978-1985), Bosnia (1991-2004), Iraq (2003-2005) and Afghanistan (2006-2010). PTSD is associated with various alterations in stress-related neurobiological systems; see box 2.

Effect of disturbed sleep on PTSD symptoms

As mentioned above, sleep is important for memory formation, under which fear extinction memory. This refers to a process of "forgetting" the association between a certain neutral trigger and an aversive stimulus. Like learning in general, fear extinction memory consolidation requires synaptic plasticity. Fear extinction memory appears to fail in patients with PTSD, who show generalization of fear responses and hyperarousal in situations that would normally be considered safe. Also in experimental studies, PTSD is related to failure of extinction (Milad et al., 2009). Remarkably, sleep, and presumably REM sleep, enhances the process of fear extinction. Triggers that were formerly related with aversive stimuli evoked lower stress responses in participants who slept following an 'extinction paradigm' (Pace-Schott et al., 2009; Spoormaker et al., 2011; Spoormaker et al., 2010). Interestingly, longitudinal studies have reported an association between sleep disturbances in the early aftermath of trauma and the development of PTSD (Koren et al., 2002; Mellman et al., 2004; Mellman et al., 2002; Kobayashi et al., 2008). Other studies also found that sleep disturbances before the trauma, as measured retrospectively, predicted PTSD (Bryant et al., 2010; Mellman et al., 1995). This indicates that sleep disturbances may indeed be related to the pathophysiology of PTSD, by for instance disrupting fear extinction memory consolidation during sleep. However, to date the effect of decreased sleep quality before trauma on PTSD development has not been studied in a prospective design.

Sleep may be fragmented by a number of external factors, such as sleep apneas, which lead to frequent short arousals during sleep. A high prevalence of obstructive sleep apnea syndrome (OSAS) has been reported in PTSD (Krakow et al., 2000; Krakow et al., 2002;

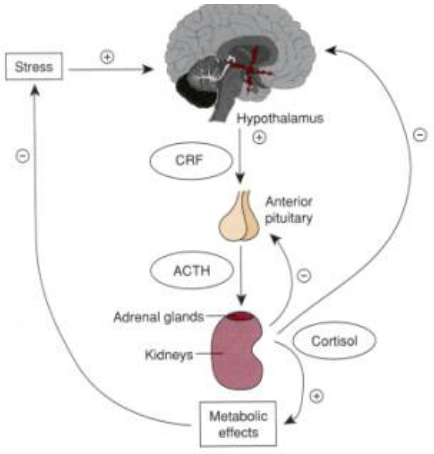
Krakow et al., 2004; Krakow et al., 2006). These studies also suggested improvement of PTSD when OSAS was successfully treated. When sleep is disturbed by obstructive airway events during sleep, followed by a short arousal and thus interrupted sleep, the decreased sleep quality may compromise beneficial processes during sleep, and may hypothetically lead to therapy resistance or a chronicity in the course of the illness.

Neurobiological underpinnings of sleep in PTSD

In patients with PTSD, activity of the hypothalamo-pituitary-adrenal (HPA)-axis and locus coeruleus/ noradreneric system is altered (see box 2), which may be responsible for over-stimulation of the amygdala and related limbic structures in the brain, as well as dysregulation of the negative feedback system that need to dampen stress responsivity (for a review see Sherin and Nemeroff, 2011).

BOX 2. Biological alterations in posttraumatic stress disorder.

Altered actvity of several brain areas has been described in patients with PTSD. Firstly, amygdala-prefrontal circuits are dysfunctional. Higher reactivity of amygdala and decreased inhibition from the medial prefrontal cortex have been reported, leading to increased fear reactions and memory flashbacks. Secondly, smaller hippocampal volume has been reported in PTSD. Also, the activity of stress hormones is altered in PTSD. Corticotrophin-releasing hormone (CRH), adrenocorticotrophic hormone (ACTH) and cortisol are hormones within the hypothalamo-pituitary-adrenal (HPA)-axis. Cortisol modulates amygdala and hippocampal functioning through glucocorticoid receptors (GR) and mineralocorticoid receptors (MR). During normal functioning of the HPA-axis, acute stress stimulates CRH release from hypothalamic neurons into the pituitary, where it stimulates ACTH secretion into the circulation. ACTH binding at the adrenals causes cortisol release into the blood. HPA-axis activity is self-limiting as cortisol inhibits CRH and ACTH secretion via negative feedback on the hypothalamus and pituitary. The HPA-axis follows a circadian rhythm, with lowest cortisol levels during the early night, and highest levels upon awakening. In PTSD, increased CRH levels with normal or decreased levels of plasma cortisol have been described, as well as a more exaggerated reaction to the GR agonist dexamethasone. Additionally, increased noradrenaline levels have been described in PTSD, which is released from the locus coeruleus (LC) in the pons.



At night, the altered activity of these neurohormones, neurotransmitters and limbic structures may lead to altered REM sleep and arousal regulation (Germain et al., 2008). The alteration in stress-related biological systems may be related to insomnia and re-experiencing of the trauma during REM sleep, when limbic structures are most active (for a review see Dang-Vu et al., 2007). The restoration of sleep in patients with PTSD may improve sleep-dependent neuroplasticity and stimulate recovery. However, only few randomized controlled trials have investigated how sleep disturbances in PTSD can be alleviated. In order to be able to improve sleep, the profile of sleep disturbances in PTSD and the underlying neurobiological systems require further study. So far, polysomnographic studies have not clarified regulating mechanisms involved in sleep disturbances in PTSD. This warrants alternative methods for the assessment of sleep regulation in PTSD.

RESEARCH QUESTIONS

This dissertation consists of several experimental studies designed to answer the following questions regarding the relationship between sleep and PTSD:

- Do insomnia and nightmares before deployment predict the development of PTSD symptoms?
- Is the prevalence of obstructive sleep apnea syndrome increased in PTSD? Are apneas related to nightmares and insomnia complaints, or to PTSD severity?
- Are heart rate during sleep and the nocturnal secretion of melatonin, cortisol and ACTH altered in PTSD, and related to decreased objective and subjective sleep quality?
- What is the relationship between objective sleep parameters according to polysomnography and subjective sleep quality in PTSD?
- Are growth hormone secretion and memory formation disturbed in PTSD, and is this due to decreased objective sleep quality?
- What is the effect of prazosin, a α_1-adrenoceptor antagonist, on objective and subjective sleep quality in PTSD when compared to a placebo?

GENERAL OUTLINE

In chapter 2 we describe a prospective longitudinal cohort study conducted on soldiers who went on a military deployment to Afghanistan (n= 453). In this study we used a regression analysis to test the predictive value of nightmares and insomnia before deployment for the development of PTSD symptoms, as assessed 6 months post-deployment. We controlled for demographic variables, early life trauma, as well as depression and anxiety scores before deployment. **In chapter 3** the prevalence of obstructive sleep ap-

nea syndrome (OSAS) was studied with polysomnography in veterans with PTSD, and age matched veterans without lifetime psychiatric disorders (trauma controls, TC), and non-trauma-exposed controls (healthy controls, HC). The relationship between OSAS and daytime symptoms in PTSD was assessed. **Chapter 4** describes our study on simultaneous polysomnography, subjective sleep measurements and nocturnal plasma levels of ACTH, cortisol and melatonin in PTSD, TCs and HCs. Results for group differences in and correlations between sleep architecture, awakenings, subjective sleep measures, heart rate and nocturnal ACTH, cortisol and melatonin will be discussed. C**hapter 5** focuses on the effect of growth hormone secretion during the night. Also, the relationship between sleep fragmentation, growth hormone secretion and sleep dependent memory formation is investigated in patients with PTSD, TCs, and HCs. **Chapter 6** comprises a systematic review regarding pharmacological interventions for sleep problems in PTSD. **Chapter 7** shows the results of a small randomized controlled trial using polysomnography to test the effect of prazosin, a α_1-adrenoceptor antagonist, versus placebo on objective and subjective sleep quality in patients with PTSD. The results of all chapters are summarized and discussed in **chapter 8**.

REFERENCE LIST

Breslau N, Roth T, Burduvali E, Kapke A, Schultz L, Roehrs T (2004) Sleep in lifetime posttraumatic stress disorder: a community-based polysomnographic study. Arch Gen Psychiatry 61:508-516.

Bryant RA, Creamer M, O'Donnell M, Silove D, McFarlane AC (2010) Sleep disturbance immediately prior to trauma predicts subsequent psychiatric disorder. Sleep 33:69-74.

Buchsbaum MS, Hazlett EA, Wu J, Bunney WE, Jr. (2001) Positron emission tomography with deoxyglucose-F18 imaging of sleep. Neuropsychopharmacology 25:S50-S56.

Cirelli C, Tononi G (2000) Gene expression in the brain across the sleep-waking cycle. Brain Res 885:303-321.

Dang-Vu TT, Desseilles M, Petit D, Mazza S, Montplaisir J, Maquet P (2007) Neuroimaging in sleep medicine. Sleep Med 8:349-372.

de Vries GJ, Olff M (2009) The lifetime prevalence of traumatic events and posttraumatic stress disorder in the Netherlands. J Trauma Stress 22:259-267.

Eriksson PS, Perfilieva E, Bjork-Eriksson T, Alborn AM, Nordborg C, Peterson DA, Gage FH (1998) Neurogenesis in the adult human hippocampus. Nat Med 4:1313-1317.

Germain A, Buysse DJ, Nofzinger E (2008) Sleep-specific mechanisms underlying posttraumatic stress disorder: integrative review and neurobiological hypotheses. Sleep Med Rev 12:185-195.

Geuze E, Vermetten E, Bremner JD (2005) MR-based in vivo hippocampal volumetrics: 2. Findings in neuropsychiatric disorders. Mol Psychiatry 10:160-184.

Hurwitz TD, Mahowald MW, Kuskowski M, Engdahl BE (1998) Polysomnographic sleep is not clinically impaired in Vietnam combat veterans with chronic posttraumatic stress disorder. Biol Psychiatry 44:1066-1073.

Johnson DR, Fontana A, Lubin H, Corn B, Rosenheck R (2004) Long-term course of treatment-seeking Vietnam veterans with posttraumatic stress disorder: mortality, clinical condition, and life satisfaction. J Nerv Ment Dis 192:35-41.

Kessler RC, McGonagle KA, Zhao S, Nelson CB, Hughes M, Eshleman S, Wittchen HU, Kendler KS (1994) Lifetime and 12-month prevalence of DSM-III-R psychiatric disorders in the United States. Results from the National Comorbidity Survey. Arch Gen Psychiatry 51:8-19.

Kobayashi I, Boarts JM, Delahanty DL (2007) Polysomnographically measured sleep abnormalities in PTSD: a meta-analytic review. Psychophysiology 44:660-669.

Kobayashi I, Sledjeski EM, Spoonster E, Fallon WF, Jr., Delahanty DL (2008) Effects of early nightmares on the development of sleep disturbances in motor vehicle accident victims. J Trauma Stress 21:548-555.

Koren D, Arnon I, Lavie P, Klein E (2002) Sleep complaints as early predictors of posttraumatic stress disorder: a 1-year prospective study of injured survivors of motor vehicle accidents. Am J Psychiatry 159:855-857.

Krakow B, Germain A, Tandberg D, Koss M, Schrader R, Hollifield M, Cheng D, Edmond T (2000) Sleep breathing and sleep movement disorders masquerading as insomnia in sexual-assault survivors. Compr Psychiatry 41:49-56.

Krakow B, Haynes PL, Warner TD, Santana E, Melendrez D, Johnston L, Hollifield M, Sisley BN, Koss M, Shafer L (2004) Nightmares, insomnia, and sleep-disordered breathing in fire evacuees seeking treatment for posttraumatic sleep disturbance. J Trauma Stress 17:257-268.

Krakow B, Melendrez D, Warner TD, Clark JO, Sisley BN, Dorin R, Harper RM, Leahigh LK, Lee SA, Sklar D, Hollifield M (2006) Signs and symptoms of sleep-disordered breathing in trauma

survivors: a matched comparison with classic sleep apnea patients. J Nerv Ment Dis 194:433-439.

Krakow B, Melendrez D, Warner TD, Dorin R, Harper R, Hollifield M (2002) To breathe, perchance to sleep: sleep-disordered breathing and chronic insomnia among trauma survivors. Sleep Breath 6:189-202.

Lewis DA, Gonzalez-Burgos G (2008) Neuroplasticity of neocortical circuits in schizophrenia. Neuropsychopharmacology 33:141-165.

Manji HK, Quiroz JA, Sporn J, Payne JL, Denicoff K, Gray A, Zarate CA, Jr., Charney DS (2003) Enhancing neuronal plasticity and cellular resilience to develop novel, improved therapeutics for difficult-to-treat depression. Biol Psychiatry 53:707-742.

Maquet P, Ruby P, Maudoux A, Albouy G, Sterpenich V, ng-Vu T, Desseilles M, Boly M, Perrin F, Peigneux P, Laureys S (2005) Human cognition during REM sleep and the activity profile within frontal and parietal cortices: a reappraisal of functional neuroimaging data. Prog Brain Res 150:219-227.

Mellman TA, Bustamante V, Fins AI, Pigeon WR, Nolan B (2002) REM sleep and the early development of posttraumatic stress disorder. Am J Psychiatry 159:1696-1701.

Mellman TA, David D, Kulick-Bell R, Hebding J, Nolan B (1995) Sleep disturbance and its relationship to psychiatric morbidity after Hurricane Andrew. Am J Psychiatry 152:1659-1663.

Mellman TA, Knorr BR, Pigeon WR, Leiter JC, Akay M (2004) Heart rate variability during sleep and the early development of posttraumatic stress disorder. Biol Psychiatry 55:953-956.

Milad MR, Pitman RK, Ellis CB, Gold AL, Shin LM, Lasko NB, Zeidan MA, Handwerger K, Orr SP, Rauch SL (2009) Neurobiological basis of failure to recall extinction memory in posttraumatic stress disorder. Biol Psychiatry 66:1075-1082.

Mueller AD, Mear RJ, Mistlberger RE (2011) Inhibition of hippocampal neurogenesis by sleep deprivation is independent of circadian disruption and melatonin suppression. Neuroscience 193:170-181.

Neylan TC, Marmar CR, Metzler TJ, Weiss DS, Zatzick DF, Delucchi KL, Wu RM, Schoenfeld FB (1998) Sleep disturbances in the Vietnam generation: findings from a nationally representative sample of male Vietnam veterans. Am J Psychiatry 155:929-933.

Neylan TC, Mueller SG, Wang Z, Metzler TJ, Lenoci M, Truran D, Marmar CR, Weiner MW, Schuff N (2010) Insomnia severity is associated with a decreased volume of the CA3/dentate gyrus hippocampal subfield. Biol Psychiatry 68:494-496.

Ohayon MM, Shapiro CM (2000) Sleep disturbances and psychiatric disorders associated with posttraumatic stress disorder in the general population. Compr Psychiatry 41:469-478.

Pace-Schott EF, Milad MR, Orr SP, Rauch SL, Stickgold R, Pitman RK (2009) Sleep promotes generalization of extinction of conditioned fear. Sleep 32:19-26.

Peigneux P, Laureys S, Fuchs S, Collette F, Perrin F, Reggers J, Phillips C, Degueldre C, Del FG, Aerts J, Luxen A, Maquet P (2004) Are spatial memories strengthened in the human hippocampus during slow wave sleep? Neuron 44:535-545.

Pillar G, Malhotra A, Lavie P (2000) Post-traumatic stress disorder and sleep-what a nightmare! Sleep Med Rev 4:183-200.

Rasch B, Buchel C, Gais S, Born J (2007a) Odor cues during slow-wave sleep prompt declarative memory consolidation. Science 315:1426-1429.

Rasch B, Dodt C, Molle M, Born J (2007b) Sleep-stage-specific regulation of plasma catecholamine concentration. Psychoneuroendocrinology 32:884-891.

Riemann D, Voderholzer U, Spiegelhalder K, Hornyak M, Buysse DJ, Nissen C, Hennig J, Perlis ML, van Elst LT, Feige B (2007) Chronic insomnia and MRI-measured hippocampal volumes: a pilot study. Sleep 30:955-958.

Sherin JE, Nemeroff CB (2011) Post-traumatic stress disorder: the neurobiological impact of psychological trauma. Dialogues Clin Neurosci 13:263-278.

Solomon Z, Mikulincer M (2006) Trajectories of PTSD: a 20-year longitudinal study. Am J Psychiatry 163:659-666.

Spoormaker VI, Schroter MS, Andrade KC, Dresler M, Kiem SA, Goya-Maldonado R, Wetter TC, Holsboer F, Samann PG, Czisch M (2011) Effects of rapid eye movement sleep deprivation on fear extinction recall and prediction error signaling. Hum Brain Mapp.

Spoormaker VI, Sturm A, Andrade KC, Schroter MS, Goya-Maldonado R, Holsboer F, Wetter TC, Samann PG, Czisch M (2010) The neural correlates and temporal sequence of the relationship between shock exposure, disturbed sleep and impaired consolidation of fear extinction. J Psychiatr Res 44:1121-1128.

Troxel WM, Kupfer DJ, Reynolds CF, III, Frank E, Thase ME, Miewald JM, Buysse DJ (2011) Insomnia and objectively measured sleep disturbances predict treatment outcome in depressed patients treated with psychotherapy or psychotherapy-pharmacotherapy combinations. J Clin Psychiatry.

Van Der Werf YD, Altena E, Schoonheim MM, Sanz-Arigita EJ, Vis JC, De RW, Van Someren EJ (2009) Sleep benefits subsequent hippocampal functioning. Nat Neurosci 12:122-123.

Wang G, Grone B, Colas D, Appelbaum L, Mourrain P (2011) Synaptic plasticity in sleep: learning, homeostasis and disease. Trends Neurosci 34:452-463.

Wilson MA, McNaughton BL (1994) Reactivation of hippocampal ensemble memories during sleep. Science 265:676-679.

Yoo SS, Hu PT, Gujar N, Jolesz FA, Walker MP (2007) A deficit in the ability to form new human memories without sleep. Nat Neurosci 10:385-392.

Chapter 2

The impact of impaired sleep as predictor of PTSD symptoms in combat veterans; a prospective longitudinal cohort study

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ABSTRACT

Background. A significant proportion of soldiers return from deployment with symptoms of fatigue, sleep difficulties and posttraumatic complaints. Disrupted sleep has been proposed as a contributing factor for the development of posttraumatic stress disorder (PTSD).

Aim. This study investigates the impact of impaired sleep and nightmares before deployment on the development of PTSD symptoms.

Methods. We collected reports on insomnia and nightmares in 453 Dutch service members prior to military deployment to Afghanistan. PTSD symptoms were assessed at 6 months post-deployment. The predictive value of insomnia complaints and nightmares on the development of PTSD symptoms was assessed with a hierarchical multiple regression analyses, in which was controlled for pre-deployment mood and anxiety symptoms, demographic variables and early life trauma.

Results. Self-reported pre-deployment insomnia complaints and nightmares predicted PTSD symptoms at 6 months. The effect of pre-existing insomnia complaints on the development of PTSD symptoms disappeared after correcting for pre-deployment mood and anxiety scores, which were significant predictors for PTSD symptoms ($p<0.05$). The effect remained for pre-deployment nightmares, which were an independent predictor of post-deployment PTSD symptoms ($p<0.05$).

Conclusion. Nightmares independently predicted PTSD symptoms, after controlling premorbid mood and anxiety complaints, however insomnia symptoms did not.

INTRODUCTION

During the last decades, large cohorts of healthy young males and females have participated in military deployment to war zones. Combat has its risk for danger to life and being exposed to enemy fire, participation in armed combat, seeing injured comrades as well as civilians, and witnessing death of fellow soldiers and civilians. Fortunately, most soldiers cope well after homecoming, but some develop deployment related illness such as posttraumatic stress disorder (PTSD) (Engelhard et al., 2007; Fear et al., 2010; Thomas et al., 2010). Despite selection, optimal training and preparation the incidence varies between 3-15 %. (Engelhard et al., 2007; Fear et al., 2010; Thomas et al., 2010) Early interventions may prevent the development of PTSD in those who are at risk. Yet, only few studies thus far reported positive effects of these early interventions. For improvement of preventive strategies it remains very important to determine who is at highest risk for developing symptoms of PTSD.

PTSD is typically characterized by unsuccessful fear extinction (Milad et al., 2009). Poor sleep after a traumatic experience has been hypothesized to contribute to the pathogenesis of PTSD, by means of disrupting the presumed beneficial process of sleep on fear extinction (Pace-Schott et al., 2009). This hypothesis has been supported by studies that observed sleep disturbances in the early aftermath of trauma in individuals who later went on to develop PTSD. Preexisting mood and anxiety disorders may also contribute to the development of PTSD, and are frequently accompanied by insomnia symptoms or nocturnal anxiety (Abad and Guilleminault, 2005). The effect of a pre-trauma psychiatric disorders on the development of trauma related psychiatric disorders was recently addressed in a study by Bryant et al (2010). However, in this study anxiety and mood complaints were not assessed at the time period in which insomnia complaints were measured. Therefore it is not sufficiently excluded that the reported relationship between sleep disturbances and development of PTSD was influenced by the effect of anxiety and mood complaints on the development of PTSD. Also early life trauma is a vulnerability factor for PTSD development (1993). In the current study we analyzed the relationship between pre-trauma sleep complaints, mood and anxiety symptoms, childhood trauma and PTSD symptoms using a prospective design. We hypothesized that insomnia complaints and nightmares prior to deployment predict PTSD symptoms at 6 months post deployment.

MATERIALS AND METHODS

Procedures

Participants were selected from a large prospective cohort study on biological and psychological effects of deployment in military personnel of the Dutch Armed Forces

(van Zuiden M. et al., 2011). Participants volunteered to participate prior to a 4-month NATO-International Security Assistance Force (ISAF) deployment to Afghanistan. Duties during deployment included combat patrols, clearing or searching buildings, participation in de-mining operations, and transportation across enemy territory. They were exposed to typical war zone stressors such as enemy fire, armed combat, and seeing deceased or seriously injured fellow soldiers and civilians and witnessing death of fellow soldiers and civilians.

Participants were recruited at military bases in the Netherlands between 2005 and 2008. Written consent was obtained from all participants after a written and verbal description of the study. After signing the informed consent, questionnaires were administered that included information on demographic variables, previous deployments and psychiatric and somatic complaints. All questionnaires were repeated at 6 months post deployment. Assessments were executed at military bases or at the Research Center of the Military Mental Health Unit at the Central Military Hospital in Utrecht, the Netherlands. The study was approved by the Institutional Review Board of the University Medical Center Utrecht, the Netherlands.

Participants

A total of 704 subjects were included. Participants were excluded from the analyses in case of missing demographic data (n = 34), missing sleep item scores (n = 3), missing pre-deployment anxiety/depression scores (n = 6), and missing post deployment assessment scores at 6 months after deployment (n = 160). Furthermore, since we were interested in development of PTSD symptoms, participants with a pre-deployment SRIP score above the cut-off of ≥ 38 (van Zuiden M. et al., 2011) were excluded from the analyses (n = 30).

Questionnaires

The Self-Rating Inventory for PTSD (SRIP; (Hovens et al., 1994) was used to assess PTSD symptoms before and at 6 months after deployment. The SRIP is a Dutch validated and reliable self-administered questionnaire for assessing PTSD symptoms in the past 4 weeks (Hovens et al., 1994). This questionnaire consists of 22 questions, ranging from 1 (never) to 4 (very frequent). A higher score indicates more PTSD symptoms (range 22- 88). The SRIP has good concurrent validity with other PTSD measures such as the Clinician Administered PTSD Scale (CAPS) and Mississippi scale for PTSD (Hovens et al., 1994). The SRIP includes three sleep questions (“I had bad dreams”, “I had difficulties falling asleep”, “I had difficulties maintaining sleep”). We also calculated a Non-sleep SRIP score without inclusion of these three sleep questions in the total score. We used this adapted SRIP score as a covariate in the regression analyses to correct for the predictive value of non-sleep PTSD symptoms before deployment on PTSD symptoms after deployment.

All participants completed the Dutch version of the Symptom Checklist (SCL-90) (Arrindell WA and Etterna JHM, 2003). The SCL-90 consists of 90 items, ranging from 1 (not at all) to 5 (very much). It is designed to measure eight subscales: anxiety, agoraphobia, depression, somatic complaints, insufficiency of thinking and acting, interpersonal sensitivity, hostility and sleeping problems. Depression and anxiety scores (range respectively 16-80 and range 10-50) were determined by the two corresponding subscales.

Sleep complaints were assessed by using three items of the SRIP ("I had bad dreams", "I had difficulties falling asleep", "I had difficulties maintaining sleep") and three items from the SCL-90 ("Difficulties falling asleep", "Waking up too early", "Restless or disturbed sleep.") The scores on the 5 questions regarding insomnia complaints (sleep initiation, sleep maintenance, early awakening and disturbed sleep) were added to construct a total score for insomnia symptoms (Cronbach's alpha 0.77). As the item scores differed in range, we calculated standard scores (z-values) for each item. Z-values of the two questions on sleep initiation were averaged and used as one z-value. Nightmare severity was measured using the SRIP item "I had bad dreams", and was analyzed independently of the constructed category of insomnia symptoms.

Traumatic experiences during childhood were determined using the Dutch short version of the Early Trauma Inventory (ETI) (Rademaker et al., 2008). The ETI consists of 27 dichotomous (yes/no) items assessing general trauma, physical abuse, emotional abuse and sexual abuse before the age of 18. The total score, which represents the sum of all experienced events, was used for further analyses.

Statistical analyses

Statistical analyses were performed using the Statistical Package for the Social Sciences version (SPSS) 15.0. Results were considered significant at $p < 0.05$. Variables were checked for normality and a log transformation was performed when appropriate. Anxiety and depression scores on the SCL-90 subscales were highly skewed. In order to acquire normally distributed variables, outliers on anxiety or depression scores (mean + 3SD) (n = 18) were excluded and Box-Cox transformations were applied by using Minitab Statistical Software. Before Box-Cox transformations were calculated, data were recoded in order to preserve the original direction of regression coefficients. The difference between SRIP scores and the insomnia symptoms before and after deployment was analyzed with a repeated measures ANOVA. Changes in depression scores, anxiety scores and nightmares over time were analyzed with the nonparametric Wilcoxon Signed Rank Test for a related sample.

In order to assess the contribution of insomnia symptoms and nightmares to PTSD symptoms post deployment, a hierarchical multiple regression analysis was performed with the total SRIP score at 6 months post deployment as dependent variable. The first block comprised insomnia symptoms and nightmares scores. Pre-deployment psychiat-

ric symptoms were entered in the second block, and demographic characteristics (age, gender, education, smoking, alcohol, rank, number of previous deployments, ETI) in the third block. Variance Inflation Factors (VIF) were reviewed to screen for multicollinearity problems. Multicollinearity was considered absent when variables had a VIF < 5 (Cohen et al., 2003).

RESULTS

Demographic and psychometric characteristics

The final sample consisted of 453 individuals, of whom demographic data are presented in table 1.

The mean SRIP score, the insomnia score and nightmare complaints increased marginally, but significantly after deployment, while depression and anxiety scores before deployment were similar to depression and anxiety scores after deployment when calculated for the whole group (n=453), of which 30 (6.6%) developed PTSD symptoms above the cut-off of 38 (See table 2).

Effect of pre-trauma sleep complaints on development of deployment- related PTSD symptoms

Hierarchical multiple regression analysis indicated that the full model explained 22.4% of variance in PTSD symptoms at 6 months post deployment, F (13,439) = 11.093, p < 0.001 (See table 3). The highest VIF value was 3,2 (for rank), thus multicollinearity was not a problem. The first block (insomnia symptoms and nightmares) contributed significantly to the model, and explained 10.9% increase in R^2, F (2,450) = 25.9, p < 0.001. Both insomnia symptoms and nightmares were significant predictors (resp. p = 0.02 and p < 0.01) (See table 2). The second block (pre-trauma mood, anxiety and PTSD symptoms) also contributed significantly to the model, with a 11.7% increase in R^2, F (5, 447) = 25.5, p < 0.001. Pre-deployment mood, anxiety and PTSD symptoms were all significant predictors. Interestingly, in the second block, the effect of insomnia symptoms on post deployment PTSD symptoms was no longer significant (p=0.79). Yet, in contrast, the contribution of nightmares remained highly significant (p<0.01). The third block (demographic variables) did not contribute to the model (ΔR^2 = 2.3%, F (13,439) = 1.665, p = 0.11).

DISCUSSION

This is the first prospective study that demonstrates that self-reported nightmares prior to exposure to combat independently predict PTSD symptoms 6 months post deployment

Table 1. Demographic characteristics.

	Mean	Standard deviation (SD)
Age	28.85	9.06
Early life trauma inventory	3.00	2.60
	Count	**Percentage**
Gender		
Male	421	92.9
Female	32	7.1
Marital status		
Married	119	26.3
Divorced	2	0.4
Single	154	34.0
Cohabitating	91	20.1
Relationship	87	19.2
Education		
Low	174	38.4
Moderate	233	51.4
High	46	10.2
Smoking (yes)	190	41.9
Alcohol		
None	47	10.4
1-20 units/week	382	84.3
> 20 units/week	24	5.3
Rank		
Officers	64	14.2
Non-Commissioned Officers	126	27.8
Corporals	85	18.8
Soldiers	178	39.3
Previously deployed		
Yes	230	50.8
No	223	49.2

Table 2a. Psychiatric symptoms pre and post-deployment in all subjects

Psychiatric symptoms (n = 453)	Before deployment		After deployment		Significance
SRIP *(M, SD)*	25.7	(3.5)	27.0	(6.0)	p = 0.002*
Insomnia *(M, SD)*	3.81	(2.0)	5.38	(2.8)	P < 0.001*
Nightmares *(M, SD)*	1.08	(0.3)	1.17	(0.5)	P < 0.001^{+}
SCL depression *(M, SD)*	17.4	(1.9)	17.9	(3.2)	p = 0.124^{+}
SCL anxiety *(M, SD)*	10.7	(1.0)	10.8	(1.8)	p = 0.742^{+}
SRIP ≥ 38 *(count, %)*		0	30	(6.6%)	

Table 2b. Psychiatric symptoms pre and post-deployment in subject with post-deployment SRIP ≥ 38

Psychiatric complaints (n = 30)	Before deployment		After deployment		Significance
SRIP *(M, SD)*	24.5	(2.8)	43.8	(5.1)	$p < 0.001$*
Insomnia *(M, SD)*	4.5	(2.4)	9.1	(3.3)	$p < 0.001$*
Nightmares *(M, SD)*	1.27	(0.5)	2.0	(0.98)	$p < 0.001$+
SCL depression *(M, SD)*	18.8	(2.7)	23.1	(4.5)	$p < 0.001$*
SCL anxiety *(M, SD)*	11.5	(1.4)	13.5	(4.1)	$p = 0.002$+

M = Mean, SD = Standard Deviation, SCL = Symptom Checklist, SRIP = Self Reported Inventory for Posttraumatic Stress Disorder

* Repeated Measures ANOVA

+ Wilcoxon signed rank test

Table 3. Linear regression on PTSD symptom severity (SRIP score) 6 months post deployment.

	Variables	β	Significance	Adjusted R^2	ΔR^2
Step 1				0.105	0.109
	Insomnia symptoms	0.105	$p = 0.02$		
	Nightmares	0.287	$p < 0.01$		
Step 2				0.217	0. 117
	Insomnia symptoms	-0.012	$p = 0.79$		
	Nightmares	0.213	$p < 0.01$		
	Anxiety pre deployment +	0.136	$p < 0.01$		
	Depression pre deployment +	0.123	$p = 0.02$		
	PTSD symptoms pre deployment (without sleep items)	0.224	$p < 0.01$		
Step 3				0.224	0.023
	Insomnia symptoms	0.008	$p = 0.86$		
	Nightmares	0.210	$p < 0.01$		
	Anxiety pre deployment +	0.131	$p < 0.01$		
	Depression pre deployment +	0.122	$p = 0.02$		
	PTSD symptoms pre deployment (without sleep items)	0.196	$p < 0.01$		
	Age	-0.025	$p = 0.73$		
	Early life trauma inventory	0.045	$p = 0.29$		
	Gender	0.047	$p = 0.29$		
	Education	0.022	$p = 0.67$		
	Smoking	0.042	$p = 0.35$		
	Alcohol	0.058	$p = 0.20$		
	Rank	-0.081	$p = 0.28$		
	No. of previous deployments	0.019	$p = 0.73$		

+ Box-Cox transformed data

PTSD = Posttraumatic Stress Disorder

and can be seen as a vulnerability factor for the development of PTSD. This study also shows that insomnia symptoms fail to predict PTSD symptoms at 6 months post deployment when pre-deployment mood and anxiety complaints are taken into account. These results contrast with previous studies in which a positive association between the development of PTSD and insomnia symptoms before or directly after trauma exposure (Koren et al., 2002; Mellman et al., 1995) was observed. This may be explained by the fact that these studies did not correct for mood and anxiety complaints. The relationship between insomnia symptoms, and mood and anxiety complaints is complex and may be bidirectional: insomnia symptoms may contribute to mood and anxiety complaints, and – vice versa - insomnia symptoms may be moderated by mood or anxiety complaints (Abad and Guilleminault, 2005). Therefore, it is possible that mood and anxiety due to insomnia increase the risk for developing PTSD after trauma, and are therefore mediators in the relationship between insomnia symptoms and PTSD development. Alternatively, mood and anxiety complaints may be confounding factors, causing both insomnia symptoms complaints and PTSD development. In the current design we could not differentiate whether mood and anxiety complaints are confounding factors or mediators in the relationship between insomnia symptoms and PTSD development.

Furthermore, our results suggest that pre-existing nightmares can be a vulnerability factor for the development of PTSD. This is also supported by two previous studies. Kobayashi et al. (2007) found that nightmares directly after trauma predicted the development of posttraumatic stress complaints. However, as reports in this study were obtained post-trauma, and nightmares are part of PTSD symptomatology, nightmares could well have been an early sign of PTSD rather than a vulnerability factor. In the study after hurricane Andrew of Mellman et al. (1995) a relationship between nightmares before trauma and PTSD symptoms after trauma has been described. In this study, reports on nightmare frequency were obtained retrospectively after the trauma occurred and are therefore subject to recollection bias. Due to the prospective design of the current study we were able to obtain reliable reports on nightmares before exposure to trauma.

There are various observations from biological studies that can help explain the contribution of nightmares on subsequent PTSD symptoms. It has been suggested that increased activity of noradrenaline during sleep may be involved in the aetiology of nightmares (Nielsen and Levin, 2007). This idea is supported by the observation that the selective α_1-adrenoceptor blocker Prazosin is effective in the treatment of nightmares in PTSD patients (Raskind et al., 2007). The noradrenergic system has also been associated with re-experiencing symptoms, fear conditioning and the risk of developing PTSD (Pitman and Delahanty, 2005; Southwick et al., 2002). Thus, it cannot be excluded that nightmares (before deployment) and the development of PTSD after deployment are epiphenomena both induced by an increased noradrenergic activity. This would imply that nightmares are not necessarily a causal factor for the development of PTSD. Nevertheless, evidence

from a previous studies showed that sleep positively affected generalization of extinction memory in healthy humans (Pace-Schott et al., 2009; Spoormaker et al., 2010; Spoormaker et al., 2011) indicating that disturbed sleep may contribute to PTSD development directly, by means or disrupting the beneficial process of sleep on fear extinction.

The results of the current study should be interpreted in light of the following considerations. A limitation is that we assessed sleep complaints with 6 questions from the SCL-90 and the SRIP, and did not use a more elaborate sleep questionnaire. As the Cronbach's alpha of the constructed insomnia symptoms score was good (0.77), we do not expect that this had a negative influence on the reliability of our results. Another limitation is that we only had access to subjective reports of insomnia symptoms and did not obtain objective sleep recordings. Therefore, we could not investigate whether decreased total sleep time, increased REM sleep or sleep fragmentation increased the risk for developing PTSD. A third limitation of this study is that we assessed PTSD symptoms with a self-administered questionnaire. The diagnosis of PTSD was not confirmed by a semi-structured interview such as the Clinician Administered Scale for PTSD (CAPS). Therefore, the effect of pre-trauma nightmares on the development of clinical significant PTSD according to DSM-IV criteria has not yet been proven in the current study. However, the SRIP is a validated and reliable questionnaire for assessing the severity of PTSD complaints, and has good concurrent validity with the CAPS (Hovens et al., 1994). A fourth limitation is that we assessed PTSD symptoms 6 months after deployment, while PTSD symptoms can develop several years post-deployment (Smid et al., 2009). Therefore we cannot rule out that participants with low SRIP scores at 6 months could develop PTSD symptoms later in their lives. Furthermore, our group existed of healthy and trained soldiers, of which 50% had been previously deployed. This raises the question whether we performed this study in a relatively resilient group of individuals. Nonetheless, 6.6 % of the individuals developed PTSD after deployment, which is similar to several other estimations of PTSD prevalence after trauma. The effect of deployment on depression, anxiety and insomnia scales was minimal when tested in the group as whole, as table 2 showed. This can be explained by the contribution of the 93.4% of subjects that did develop psychopathology. The 30 participants that developed PTDS complaints after deployment did exhibit increased scores of depression, anxiety and PTSD severity after deployment.

Assessment of sleep difficulties is a simple way of detecting a group of soldiers that is at risk for developing PTSD symptoms. If increased noradrenergic activity is related to both nightmares and PTSD, inhibition of noradrenergic activity before and directly after trauma in subjects with nightmares could decrease the risk for developing PTSD. Future research could provide treatment with pharmacotherapeutic compounds such as prazosin or propranolol (Shad et al., 2011) to dampen the noradrenergic hyperactivity prior to or during military deployment.

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REFERENCE LIST

Abad VC, Guilleminault C (2005). Sleep and psychiatry. Dialogues Clin Neurosci 7:291-303.

Arrindell WA, Etterna JHM (2003). Symptom Checklist. Handleiding bij een multidimensionele psychopathologie-indicator [Symptom Checklist: Manual for a Multidimensional Indicator of Psychopathology]. Lisse, The Netherlands: Swets & Zeitlinger.

Bremner JD, Southwick SM, Johnson DR, Yehuda R, Charney DS (1993). Childhood physical abuse and combat-related posttraumatic stress disorder in Vietnam veterans. Am J Psychiatry 150:235-239.

Bryant RA, Creamer M, O'Donnell M, Silove D, McFarlane AC (2010). Sleep disturbance immediately prior to trauma predicts subsequent psychiatric disorder. Sleep 33:69-74.

Cohen P, Cohen J, West S, Aiken L (2003). Applied multiple regression/correlation analyses for the behavioral sciences. Hilsdale, NJ: Lawrence Erlbaum.

Engelhard IM, van den Hout MA, Weerts J, Arntz A, Hox JJ, McNally RJ (2007). Deployment-related stress and trauma in Dutch soldiers returning from Iraq. Prospective study. Br J Psychiatry 191:140-145.

Fear NT, Jones M, Murphy D, Hull L, Iversen AC, Coker B, Machell L, Sundin J, Woodhead C, Jones N, Greenberg N, Landau S, Dandeker C, Rona RJ, Hotopf M, Wessely S (2010). What are the consequences of deployment to Iraq and Afghanistan on the mental health of the UK armed forces? A cohort study. Lancet 375:1783-1797.

Hovens JE, van der Ploeg HM, Bramsen I, Klaarenbeek MT, Schreuder JN, Rivero VV (1994). The development of the Self-Rating Inventory for Posttraumatic Stress Disorder. Acta Psychiatr Scand 90:172-183.

Koren D, Arnon I, Lavie P, Klein E (2002). Sleep complaints as early predictors of posttraumatic stress disorder: a 1-year prospective study of injured survivors of motor vehicle accidents. Am J Psychiatry 159:855-857.

Mellman TA, David D, Kulick-Bell R, Hebding J, Nolan B (1995). Sleep disturbance and its relationship to psychiatric morbidity after Hurricane Andrew. Am J Psychiatry 152:1659-1663.

Milad MR, Pitman RK, Ellis CB, Gold AL, Shin LM, Lasko NB, Zeidan MA, Handwerger K, Orr SP, Rauch SL (2009). Neurobiological basis of failure to recall extinction memory in posttraumatic stress disorder. Biol Psychiatry 66:1075-1082.

Nielsen T, Levin R (2007). Nightmares: a new neurocognitive model. Sleep Med Rev 11:295-310.

Pace-Schott EF, Milad MR, Orr SP, Rauch SL, Stickgold R, Pitman RK (2009b). Sleep promotes generalization of extinction of conditioned fear. Sleep 32:19-26.

Pitman RK, Delahanty DL (2005). Conceptually driven pharmacologic approaches to acute trauma. CNS Spectr 10:99-106.

Raskind MA, Peskind ER, Hoff DJ, Hart KL, Holmes HA, Warren D, Shofer J, O'Connell J, Taylor F, Gross C, Rohde K, McFall ME (2007). A parallel group placebo controlled study of prazosin for trauma nightmares and sleep disturbance in combat veterans with post-traumatic stress disorder. Biol Psychiatry 61:928-934.

Shad MU, Suris AM, North CS (2011). Novel combination strategy to optimize treatment for PTSD. Hum Psychopharmacol.

Smid GE, Mooren TT, van der Mast RC, Gersons BP, Kleber RJ (2009). Delayed posttraumatic stress disorder: systematic review, meta-analysis, and meta-regression analysis of prospective studies. J Clin Psychiatry 70:1572-1582.

Southwick SM, Davis M, Horner B, Cahill L, Morgan CA, III, Gold PE, Bremner JD, Charney DC (2002). Relationship of enhanced norepinephrine activity during memory consolidation to enhanced long-term memory in humans. Am J Psychiatry 159:1420-1422.

Spoormaker VI, Schroter MS, Andrade KC, Dresler M, Kiem SA, Goya-Maldonado R, Wetter TC, Holsboer F, Samann PG, Czisch M (2011). Effects of rapid eye movement sleep deprivation on fear extinction recall and prediction error signaling. Hum Brain Mapp.

Spoormaker VI, Sturm A, Andrade KC, Schroter MS, Goya-Maldonado R, Holsboer F, Wetter TC, Samann PG, Czisch M (2010). The neural correlates and temporal sequence of the relationship between shock exposure, disturbed sleep and impaired consolidation of fear extinction. J Psychiatr Res 44:1121-1128.

Thomas JL, Wilk JE, Riviere LA, McGurk D, Castro CA, Hoge CW (2010). Prevalence of mental health problems and functional impairment among active component and National Guard soldiers 3 and 12 months following combat in Iraq. Arch Gen Psychiatry 67:614-623.

van Zuiden M, Geuze E, Willemen HL, Vermetten E, Maas M, Heijnen CJ, Kavelaars A (2011). Pre-existing high glucocorticoid receptor number predicting development of posttraumatic stress symptoms after military deployment. Am J Psychiatry 168:89-96.

Chapter 3

Obstructive Sleep Apnea in Combat-related Post-traumatic Stress Disorder; a controlled polysomnography study

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ABSTRACT

Background. Obstructive sleep apnea (OSA) may be highly prevalent in post-traumatic stress disorder (PTSD) and may exacerbate PTSD complaints.

Aim. Our objective was to determine whether the occurence of OSA was highly prevalent in a sample of Dutch veterans with PTSD as compared with age and trauma matched controls, and whether OSA was associated with more severe PTSD complaints.

Methods. We determined the apnea hypopnea indices (AHI) with polysomnographic registrations in 20 veterans with PTSD, 24 veterans without PTSD and 17 healthy controls. PTSD severity and nightmare complaints were assessed with the Clinician Administered PTSD Scale (CAPS).

Results. The prevalence of an AHI > 10 was 29% in PTSD, 21% in TC and 29% in HCs (χ^2= 0.60, *df* = 2, *p* = n.s.). The mean CAPS score in patients with OSA (*n* = 6) was significantly higher than in patients without OSA ($p < 0.05$), while nightmare severity was similar in PTSD patients with OSA as compared with PTSD patients without OSA (*p* = n.s.). Furthermore, there was a significant correlation between AHI and CAPS score in PTSD patients ($r = 0.46$, $p < 0.05$, *df* = 14).

Conclusions. Our results indicate that PTSD is not necessarily associated with a higher prevalence of OSA. However, PTSD severity was related to OSA, which may possibly mean that co morbid OSA leads to an increase of PTSD complaints. However, future research should indicate whether OSA exerts a negative influence on PTSD, and whether treatment of OSA is alleviates PTSD symptoms.

INTRODUCTION

Traumatic memories are frequently experienced during nightmares in patients suffering from post-traumatic stress disorder (PTSD). PTSD patients are vigilant at night and complain of nightmares, frequent awakenings and non-restorative sleep (Neylan et al, 1998). Despite often severe subjective complaints, objective sleep measures, such as total sleep time and the amount REM sleep, are generally unaffected in PTSD. More subtle changes in polysomnographic recordings, such as increased stage 1 sleep, decreased REM density, and a higher number of awakenings, have been reported (Kobayashi et al., 2007; van Liempt et al., 2011), however, it is unclear how these alterations relate to insomnia and nightmares. Mechanisms responsible for the nightly complaints remain to be further explored (Spoormaker & Montgomery, 2010).

Earlier research has shown that posttraumatic nightmares and other posttraumatic complaints may be related to obstructive sleep apnea (OSA) (Krakow et al, 2000, 2001a; Yesavage et al, 2010). When OSA is highly prevalent in PTSD and related to PTSD symptoms, reducing respiratory events may be a putative focus for the treatment of posttraumatic complaints (Hurwitz & Khawaja, 2010). Thus far, only uncontrolled studies reported on a high prevalence of OSA in PTSD.

In order to examine the putative relationship between PTSD and OSA, we determined the apnea hypopnea index (AHI), Body Mass index (BMI), PTSD severity and nightmare complaints in veterans with PTSD, veterans without PTSD and healthy controls.

MATERIALS AND METHODS

Participants

Twenty-three male veterans with PTSD were recruited through the outpatient clinic of the Military Mental Healthcare (MMH), Utrecht, the Netherlands. PTSD patients with habitual benzodiazepine usage (n=1) and substance abuse were excluded (n=2). Patients who used benzodiazepines, alcohol or drugs weekly or monthly were instructed to refrain from these substances on the day of the sleep recordings. Five PTSD patients used a selective serotonin reuptake inhibitor.

Twenty-four trauma controls (TCs; veterans without PTSD), and 17 healthy controls (HCs; civilians or service members who were naïve for deployment) were recruited through advertisements in veteran related magazines and newspapers. TCs were matched for age, year and region of deployment with the PTSD group, and were excluded when they had a CAPS score of 18 or higher. HCs were matched for age with the PTSD group, and were excluded when reporting significant psychotrauma in the past according to the CAPS.

All control subjects were medically healthy males without a history of psychiatric disorders, and without sleep complaints. Written consent was obtained from all participants, after a complete written and verbal description of the study. The study was approved by the Institutional Review Board of the University Medical Centre of Utrecht, The Netherlands.

Procedures

All veterans were screened for psychiatric illness using the structured clinical interview for DSM IV axis I disorders (SCID) (Spitzer et al, 1992). The diagnosis of PTSD (assessed with the SCID) was confirmed by the Clinician Administered PTSD Scale (CAPS) (Blake et al, 1995) and after consensus by two clinicians (SvL, EV). Only patients with a CAPS score above 50 were included. Trauma controls were included if they met the A1 criterion for PTSD (the person experienced, witnessed or was confronted with an event or events that involved actual or threatened death or serious injury, or a threat to physical integrity of self or others), but had a CAPS score below 18 and did not meet DSM IV criteria for PTSD or any other life time axis I disorder. Subjects were screened for medical conditions by history taking and physical examination. As a high Body Mass Index (BMI) is associated with AHI, weight and length was determined.

PSG recordings were obtained during 1 night at Kempenhaeghe, the Netherlands, with Brainlab Real Time ©, including EOG for vertical and horizontal eye movements, EMG (chin, left and right m. tibialis anterior), ECG and EEG (F0-C0, F3-C3, P3-O1, C4-A1, O2-A2), airflow at the nose (nasal pressure), movement of the thorax and abdomen by inductance plethysmography, and arterial oxygen saturation at the index finger.

Data analyses

Sleep data were analyzed according to criteria of Rechtschaffen and Kales (1968) by an experienced sleep technician who was blind to group identity. Apneas and hypopneas were scored when an airflow reduction of 50-90 % (hypopnea) or >90 % (apnea) was detected for at least 10 seconds, except when such airflow reductions occurred after an arousals or during wake.

An AHI > 15 per hour is the cut-off for obstructive sleep apnea syndrome (OSAS) (Epstein et al, 2009). An AHI > 10 is also considered clinically relevant and used as an exclusion criterion in sleep studies (Mellman et al, 1997, Engdahl et al, 2000). We compared both indices between groups. In comparisons of mean CAPS scores between PTSD patients with and without OSA we used the cut-off of AHI > 10.

Statistical analyses

Differences between the groups were tested in SPSS 17.0. An ANOVA was used to test group differences when variables were normally distributed. In other cases a non-parametric Mann-Whitney test was used. With a Chi^2 test we examined whether OSA was more

prevalent in PTSD compared with controls. Correlations were analyzed with a Pearson's test, and were controlled for age and BMI. Differences were considered significant when *p*-values were smaller than 0.05.

RESULTS

Demographic data

In table 1, demographic variables are shown. PTSD Patients (*n* = 20), TCs (*n* = 24) and HCs (*n* = 17) did not differ in age or alcohol intake per week. The mean CAPS score was 67.50 (SD = 11.01) in PTSD patients and 4.00 (SD = 5.12) in TCs. Of 2 PTSD patients, 4 TCs and 1 HC BMI data were missing. PTSD patients (*n* = 18) had a higher BMI than both TCs (*n* = 20) and HCs (*n* = 16) (respectively 27.86 (SD = 4.86); 24.64 (SD = 2.90) and 23.65 (SD = 2.42), ANOVA F (2,51)= 6.66, $p < 0.01$.

Prevalence OSA

The AHI was higher than 15 in 10% of the PTSD patients, 13% of the TCs and 12% of the HCs ($\chi^2 = 0.07$, $df = 2$, p= n.s.). Twenty-nine percent of the PTSD patients, 21% of TC and 29% of HC had an AHI >10 per hour ($\chi^2 = 0.60$, $df = 2$, p = n.s.). The mean AHI was 6.79 (SD = 5.58) in PTSD patients, 7.18 (SD = 6.50) in TCs, and 5.68 (5.91) in HCs (ANOVA, $F\ (2,58) = 0.32$, p = n.s.).

OSA and PTSD severity

The patients with an AHI > 10 (*n* = 6) exhibited significantly higher CAPS scores (M = 77, SD = 9) than patients without OSA (M = 63, SD = 9) (*n* = 14) (Mann-Whitney U-test: z= -2.36, $p < 0.05$).

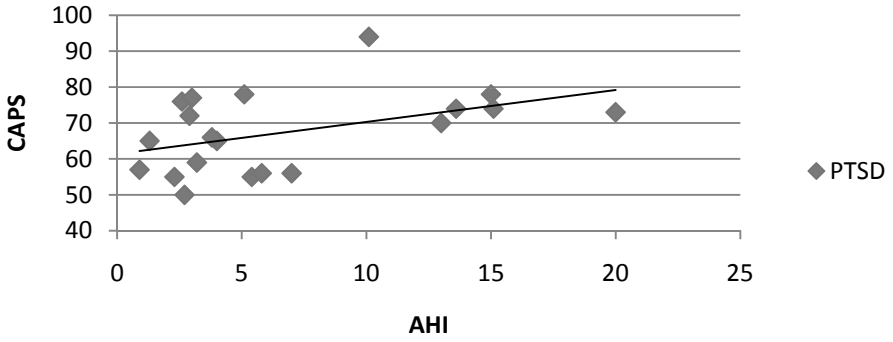
Table 1. Demographic characteristics

	PTSD (n=20)		TC (n=24)		HC (n=17)		Comparison
Age *(M, SD)*	40.75	(8.45)	37.71	(6.91)	35.06	(8.26)	$df = 60$, $F = 2.5$, $p = 0.095$
Alcohol intake *(M, SD)*	7.50	(8.00)	5.33	(7.07)	7.29	(5.28)	$df = 60$, $F = 0.96$, $p = 0.391$
BMI *(M, SD)*	27.86	(4.86)	24.64	(2.90)	23.65	(2.42)	$df = 53$, $F = 6.7$, $p = 0.003$*
CAPS *(M, SD)*	67.50	(11.02)	4.00	(5.11)	0	(0)	$df = 60$, F= 571.0, $p < 0.001$
Number of A1 trauma *(M, SD)*	8.92	(2.18)	7.24	(2.91)	1.93	(1.49)	$df = 60$, $F = 36.3$, $p < 0.001$**

BMI = Body Mass Index, HC = Healthy Controls, M = Mean, PTSD = Posttraumatic Stress Disorder, SD = Standard Deviation, TC = Trauma Controls.
*PTSD >TC, $p = 0.036$. PTSD > HC, $p = 0.006$. TC = HC, $p = 1.0$.
**PTSD =TC, $p = 0.16$. HC < PTSD, $p < 0.001$. HC < TC, $p < 0.001$.

With a partial correlation, controlling for age and BMI, a significant relationship between the CAPS and AHI was found in PTSD patients (Pearson, $r = 0.46$, $p < 0.05$, $df = 14$). (See figure 1). The 3 subscales (B,C,D) of the CAPS were not significantly related to the AHI in PTSD. However the correlation between the D-subscale (hyperarousal) and AHI was at trend-level significance in PTSD patients (Pearson, $r = 0.36$, $p = 0.08$, $df = 14$).

Figure 1. Relationship between Apnea Hypopnea Index and PTSD severity.



AHI = Apnea Hypopnea Index, CAPS = Clinician Administered PTSD Scale, PTSD = Posttraumatic Stress Disorder.

OSA and Nightmares

The mean nightmare score was 3.33 (SD = 2.56) in patients with OSA (AHI > 10, $n = 6$) and 3.21 (SD = 2.29) in patients without OSA ($n = 14$) (ANOVA, $F(1,19) = 0.1$, $p =$ n.s.). Similarly, there was no relationship between B2 score of the CAPS and AHI, when corrected for BMI and age (Pearson's test $r = -0.076$, $p =$ n.s, $df = 14$).

DISCUSSION

Our study shows that the occurrence of OSA in PTSD patients was 10% and was not increased compared with TCs and HCs, despite a higher BMI of PTSD patients. Furthermore, we found a relationship between PTSD severity and the AHI, while nightmare frequency was not related to the occurrence of apneas. The observations from our study contrast with previous reports. Several studies reported high indices of AHI in 69-91% of the PTSD patients (Krakow et al, 2001b, 2002, 2004, Yesavage et al, 2010). In these studies screening instruments for detecting OSA may have been more sensitive than in our study, especially since some studies defined a cut-off of 5 events per hour. As none of the previous studies included a control group, it cannot be concluded that the incidence of OSA is elevated in PTSD. Another explanation for the high incidence of OSA in some previous studies is that the usage of benzodiazepines was not discontinued before sleep

recordings, which increases the occurrence of OSA (Dolly & Block, 1982). In our study, participants with regular benzodiazepine usage were excluded, and participants with habitual benzodiazepine usage refrained from sleep medication in the sleep laboratory. Lastly, our study group consisted of middle-aged veterans, while other studies included predominantly female PTSD patients. The incidence of OSA may be different in other populations with PTSD. Our study underlines the importance of controlled studies to determine whether OSA is more prevalent in PTSD than in matched controls.

While controlled studies have not been published on this subject before, our results are in concordance with two sleep studies in PTSD patients that excluded an equal amount of patients and controls due to OSA (Breslau et al., 2004, Mellman et al., 1997). In contrast, Engdahl et al (2000) reported that in a sample of elderly war veterans, more PTSD patients were excluded due to OSA than controls.

In our study, PTSD severity was related to OSA. Furthermore, PTSD patients with an AHI > 10 exhibited significantly higher CAPS scores compared with PTSD patients without OSA. This may be due to decreased concentration, depression and irritability, which are common complaints in OSA (Saunamäki & Jehkonen, 2007). Also previous studies have suggested a relationship between PTSD complaints and the occurrence of OSA. In the study of Engdahl et al (2000) 4 PTSD patients with OSA improved on overall wellbeing after treatment with continuous positive airway pressure (CPAP). The relationship between remitted OSA and improvement of PTSD severity was also found in an uncontrolled retrospective study on CPAP treatment in PTSD patients (Krakow et al, 2000).

Our results did not find a relationship between nightmare frequency and OSA. This contrasts with a study in which breathing interruptions, as witnessed by bed partners, were related to nightmare frequency in PTSD patients (de Groen et al, 1993). Reports from the bed partner are less reliable for detecting OSA than polysomnography, which was used in the current study. Possibly, bed partners of patients with nightmares are more alert during the night, and therefore better aware of breathing interruptions. Therefore, breathing interruptions may be more frequently reported in patients with nightmares in the study of de Groen et al (1993), in the absence of a relationship between OSA and nightmares.

A limitation of our study is the small sample size. The incidence of OSA in PTSD patients was 10% in our study, however this cannot be generalized. Furthermore, the power for detecting correlations between OSA and symptom clusters of PTSD may have been too low. Our study consisted of a homogeneous group of middle-aged male veterans. The advantage was that the control groups were well matched. However, the results may not be extrapolated to other PTSD populations.

In summary, our results indicate that PTSD is not necessarily associated with the occurrence of OSA, as some uncontrolled studies suggested. Furthermore, OSA was not related to sleep complaints, while patients with co morbid OSA exhibited higher CAPS scores. Larger controlled studies in different populations need to be performed to provide more

reliable estimations of the incidence of OSA in PTSD. As co morbid OSA in PTSD patients may possibly lead to an increase of PTSD symptoms, the influence of OSA, and treatment of OSA, should be further explored, especially in those with refractory complaints to conventional therapeutic strategies.

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REFERENCE LIST

Blake, D.D., Weathers, F.W., Nagy, L.M., Kaloupek, D.G., Gusman, F.D., Charney, D.S.,et al. (1995). The development of a Clinician-Administered PTSD Scale. *Journal of Traumatic Stress*, 8, 75-90.

Breslau, N., Roth, T., Burduvali, E., Kapke, A., Schultz, L., & Roehrs, T. (2004). Sleep in lifetime post-traumatic stress disorder: a community-based polysomnographic study. *Archives of General Psychiatry,* 61, 508-516.

de Groen J.H., Op Den Velde W., Hovens J.E., Falger, P.R., Schouten, E.G., & Van Duijn, H. (1993) Snoring and anxiety dreams. *Sleep,* 16, 35-36.

Dolly, F.R., & Block, A.J. (1982). Effect of flurazepam on sleep-disordered breathing and nocturnal oxygen desaturation in asymptomatic subjects. *American Journal of Medicine*, 73, 239-243.

Engdahl, B.E., Eberly, R.E., Hurwitz, T.D., Mahowald, M.W., & Blake, J. (2000). Sleep in a community sample of elderly war veterans with and without posttraumatic stress disorder. *Biological Psychiatry,* 47, 520-525.

Epstein, L.J., Kristo, D., Strollo, P.J., Friedman, N., Malhotra, A., Patil, S.P., et a. (2009). Clinical Guideline for the Evaluation, Management and Long-term Care of Obstructive Sleep Apnea in Adults. *Journal of Clinical Sleep Medicine*, *5,* 263-276.

Hurwitz, T.D., & Khawaja, I. (2010). Treatment of Obstructive Sleep Apnea may be an Important Adjunct to Therapy of Posttraumatic Stress Disorder not to be Overlooked. *Sleep*, 33, 1435-1436.

Kobayashi, I., Boarts, J.M., & Delahanty, D.L. (2007). Polysomnographically measured sleep abnormalities in PTSD: a meta-analytic review. *Psychophysiology,* 44, 660-669.

Krakow, B., Lowry, C., Germain, A., Gaddy, L., Hollifield, M., Koss, M., et al. (2000). A retrospective study on improvements in nightmares and post-traumatic stress disorder following treatment for co-morbid sleep-disordered breathing. *Journal of Psychosomatic Research*, 49, 291-298.

Krakow, B., Germain, A., Warner, T.D., Schrader, R., Koss, M., Hollifield, M., et al. (2001a). The relationship of sleep quality and posttraumatic stress to potential sleep disorders in sexual assault survivors with nightmares, insomnia, and PTSD. *Journal of Traumatic Stress*, 14, 647-665.

Krakow, B., Melendrez, D., Pedersen, B., Johnston, L., Hollifield, M., Germain, A., et al. (2001b). Complex insomnia: insomnia and sleep-disordered breathing in a consecutive series of crime victims with nightmares and PTSD. *Biological Psychiatry*, 49, 948-953.

Krakow, B., Melendrez, D., Johnston, L., Warner, T.D., Clark, J.O., Pacheco, M., et al. (2002). Sleep-disordered breathing, psychiatric distress, and quality of life impairment in sexual assault survivors. *Journal of Nervous and Mental Disease*, 190, 442-452.

Krakow, B., Haynes, P.L., Warner, T.D., Santana, E., Melendrez, D., Johnston, L., et al. (2004) Nightmares, insomnia, and sleep-disordered breathing in fire evacuees seeking treatment for posttraumatic sleep disturbance. *Journal of Traumatic Stress,* 17, 257-268.

Mellman, T.A., Nolan, B., Hebding, J., Kulick-Bell, R., & Dominguez, R.A. (1997). Polysomnographic comparison of veterans with combat-related PTSD, depressed men, and non-ill controls. *Sleep*, 20, 46-51.

Neylan, T.C., Marmar, C.R., Metzler, T.J., Weiss, D.S., Zatzick, D.F., Delucchi, K.L., et al. (1998). Sleep disturbances in the Vietnam generation: findings from a nationally representative sample of male Vietnam veterans. *American Journal of Psychiatry*, 155, 929-933.

Rechtschaffen, A., & Kales, A. (1968). *A manual of Standardized Terminology, Techniques and Scoring System for Sleep Stages of Human Subjects.* Washington DC: US Government Printing Office.

Saunamäki, T., & Jehkonen, M. (2007). Depression and anxiety in obstructive sleep apnea syndrome: a review. *Acta Neurologica Scandinavia,* 116(5), 277-88.

Spitzer, R.L., Williams, J.B., Gibbon, M., & First, M.B. (1992). The Structured Clinical Interview for DSM-III-R (SCID). I: History, rationale, and description. *Archives of General Psychiatry,* 49, 624-629.

Spoormaker, V.I., & Montgomery, P. (2008). Disturbed sleep in post-traumatic stress disorder: secondary symptom or core feature? *Sleep Medicine Reviews,* 12(3), 169-84.

van Liempt, S., Vermetten, E., Lentjes, E., Arends, J., & Westenberg, H. (2011). Decreased nocturnal growth hormone secretion and sleep fragmentation in combat-related posttraumatic stress disorder; potential predictors of impaired memory consolidation. *Psychoneuroendocrinology*, 36(9), 1361-9.

Yesavage, J.A., Kinoshita, L.M., Kimball, T., Zeitzer, J., Friedman, L., Noda, A., et al. (2010). Sleep-Disordered Breathing in Vietnam Veterans with Posttraumatic Stress Disorder. *American Journal of Geriatric Psychiatry*. [Epub ahead of print]

Chapter 4

Sympathetic activity and hypothalamo-pituitary-adrenal axis activity during sleep in post-traumatic stress disorder;

a study assessing polysomnography with simultaneous blood sampling

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ABSTRACT

Background. Nightmares and insomnia in PTSD are hallmark symptoms, yet poorly understood in comparison to the advances towards a biological framework for the disorder. According to polysomnography (PSG), only minor changes in sleep architecture were described. This warrants alternative methods for assessing sleep regulation in PTSD.

Methods. After screening for obstructive sleep apnea and period limb movement disorder, veterans with PTSD (n=13), trauma controls (TCs, n=17) and healthy controls (HCs, n=15) slept in our sleep laboratory on two consecutive nights with an IV catheter out of which blood was sampled every 20 min from 2200h to 0800h. Nocturnal levels of plasma adrenocorticotropic hormone (ACTH), cortisol, melatonin were assessed in conjunction with PSG registration, as well as subjective sleep parameters.

Results. PTSD patients showed a significant increase in awakenings during sleep in comparison to both control groups. These awakenings were correlated with ACTH levels during the night, and with the subjective perception of sleep depth. Also, heart rate (HR) was significantly increased in PTSD patients as compared with both control groups. The diurnal regulation of ACTH, cortisol and melatonin appeared undisturbed. PTSD patients exhibited lower cortisol levels at borderline significance (p=0.056) during the first half of the night. ACTH levels and cortisol levels during the first half of the night were inversely related to slow wave sleep (SWS).

Conclusion. This study suggests that hypothalamo-pituitary adrenal (HPA) axis activity is related to sleep fragmentation in PTSD. Also, activity of the sympathetic nervous system (SNS) is increased during sleep in PTSD. Further research is necessary to explore the potential causal relationship between sleep problems and the activity of the HPA-axis and SNS in PTSD.

INTRODUCTION

Patients with a post-traumatic stress disorder (PTSD) frequently suffer from sleep disturbances such as nightmares, frequent awakening, and sleep initiation and sleep maintenance insomnia (Neylan et al., 1998; Ohayon and Shapiro, 2000). Sleep disturbances are of clinical relevance in PTSD, as they are often therapy-resistant (Zayfert and DeViva, 2004; Davidson et al., 2001) and related to suicide risk (Ribeiro et al., 2011). Insomnia and nightmares may also exert a negative effect on daytime symptoms (Belleville et al., 2009). Recent studies show that sleep quality may even be directly related to hippocampus functioning and morphology (Van Der Werf et al., 2009; Neylan et al., 2010). Furthermore, sleep fragmentation is related to blunted growth hormone secretion in PTSD (van Liempt et al., 2011), which may be an underlying mechanism for compromised neuroplasticity and hippocampal functioning (Kim et al., 2010).

Restoring sleep disturbances in PTSD may hypothetically stimulate neuroplasticity and recovery. In order to be able to improve sleep, we need more knowledge of the characteristics and contributing factors of sleep disturbances in PTSD. A major difficulty in research into PTSD-related sleep disturbances is that despite very frequent complaints of insomnia and nightmares, objective sleep is only mildly disturbed, according to polysomnography (PSG), which is the golden standard in sleep research (Hurwitz et al., 1998; Pillar et al., 2000). Earlier PSG studies have shown a small increase in stage 1 non rapid eye movement sleep (NREM) and decrease in slow wave sleep (SWS) (Kobayashi et al., 2007). Objective measures of total sleep time (TST), waking after sleep onset (WASO), and sleep onset latency (SOL) were not related to the subjective estimates of TST, WASO and SOL (Woodward et al., 1996). Further research is needed to explain why PTSD patients suffer from insomnia and nightmares, while their sleep architecture is mostly undisturbed with normal amounts of TST and rapid eye movement (REM) sleep.

Some studies have found increased awakenings during sleep in PTSD (Breslau et al., 2004; Habukawa et al., 2007; Mellman et al., 1995). This type of sleep fragmentation may interrupt beneficial processes during sleep, and may induce alterations in stress hormones (Steiger, 2007).

To date, little is known about the biological systems that are involved in sleep dysregulation in PTSD. The hypothalamo-pituitary adrenal (HPA) axis and the sympathetic Nervous System (SNS) are well characterized in PTSD, and have been associated with sleep regulation as well (Saper et al., 2005). Furthermore, a relationship between nightmares and circadian rhythm has been suggested (Nielsen, 2010). To the best of our knowledge, circadian rhythm disturbance have not been studied in PTSD.

In order to further unravel the profile and underlying mechanisms of sleep alteration in PTSD, we undertook a sleep study in PTSD patients that assessed three sets of parameters: 1. HPA-axis activity and melatonin secretion; 2. objective sleep structure according to

PSG, including awakenings and heart rate (HR); 3. subjective perception of sleep. Most previously conducted PSG studies used either trauma controls (TCs), or healthy controls (HCs), or a mixed group in their study design. However, studies have shown that alterations in HPA-axis functioning have also been described in trauma exposed mentally healthy individuals (de Kloet et al., 2007; Klaassens et al., 2010). We therefore included two control groups: one group of veterans with similar exposure to combat, and one group of non-combat-exposed military personnel and non-trauma-exposed civilians.

MATERIALS AND METHODS

Participants

Veterans with PTSD were recruited through the outpatient clinic of the Military Mental Healthcare (MMH), Utrecht, the Netherlands. TCs were veterans without PTSD. HCs were civilians or service members who had never been deployed. All control subjects were recruited through advertisements. Controls were matched with the PTSD group for age, year of deployment (TC) and region of deployment (TC). After a verbal and written description of the study, written informed consent was obtained. All participants were screened for psychiatric illness using the Structured Clinical Interview for DSM-IV axis I disorders (SCID-I) (Spitzer et al., 1992). The diagnosis of PTSD was confirmed by the Clinician Administered PTSD Scale (CAPS) (Blake et al., 1995), and consensus by two clinicians (SvL, EV). PTSD patients were included when they met a CAPS score of 50 and did not meet DSM-IV criteria for psychotic disorder or substance abuse (according to the SCID-I). TCs were included when they met the A1 criteria for PTSD (the person experienced, witnessed or was confronted with an event or events that involved actual or threatened death or serious injury, or a threat to physical integrity of self or others), with a CAPS score below 18. Furthermore, TCs did not meet DSM-IV criteria for any current or lifetime DSM-IV axis I disorder. Typically, TCs and PTSD patients were deployed to Afghanistan, Bosnia or Lebanon for a period of 4 to 6 months. During deployment they were exposed to war zone stressors such as enemy fire, armed combat, and seeing seriously injured fellow soldiers and civilians. HCs were screened for A1 criteria and were excluded when they had ever experienced or witnessed life-threatening events. All participants were medically healthy, male, and free from psychotropic medication and alcohol or drugs dependence in the past six months. Three PTSD patients used a beta-blocker (n=3), of whom 2 also used a calcium channel antagonist and one also a ACE inhibitor. All control subjects were without a history of psychiatric disorders, and without sleep complaints. Participants who screened positive for obstructive sleep apnea syndrome (OSAS) and periodic limb movement disorder (PLMD), according to one night of polysomnography (PSG), were excluded from the study. Eligible participants (PTSD n=13, TCs n=17, HCs n=15) were scheduled for 2

nights PSG with simultaneous blood sampling. The study was approved by the Institutional Review Board of the University Medical Centre of Utrecht, The Netherlands.

Procedures

Screening OSAS and PLMD

During one night prior to study entry PSG recordings were made with Brainlab Real Time©, including EOG for vertical and horizontal eye movements, EMG (chin, left and right m. tibialis anterior), ECG and EEG (F0-CO, F3-C3, P3-O1, C4-A1, O2-A2), airflow at the nose (nasal pressure), movement of the thorax and abdomen (by inductance plethysmography), and arterial oxygen saturation at the index finger. Apneas and hypopneas were scored when an airflow reduction of 50-90 % (hypopnea) or >90 % (apnea) was detected for at least 10 seconds, and were deleted when occurring after arousals or during wake. OSAS was defined as more than 15 apnea or hypopnea per hour. PLMD was defined as more than five PLM related arousals per hour. These subjects were excluded from participation.

Sleep registrations with simultaneous blood sampling

Participants were requested to maintain regular sleep schedules during the two weeks preceding sleep registrations. On the day prior to sleep registration, participants were instructed to refrain from alcohol and to refrain from caffeine containing beverages after 1600h. Participants slept two consecutive nights at the sleep laboratory of the Central Military Hospital in Utrecht, the Netherlands. They arrived in the sleep laboratory between 1900h and 2000h. We adjusted the 8 hour polysomnography registration to the habitual bed times of participants, with a time of lights out between 2200h and 0000h, and lights on between 0600h and 0800h.

Sleep registrations were acquired with Twente Medical Systems, Porti 7 (TMS International BV), including bipolar derivations of EMG (chin), EOG for vertical and horizontal eye movements, EEG (F0-CO, F3-C3, P3-O1, C4-A1, O2-A2) and ECG. All participants slept with an IV catheter in a fore arm vein, which was connected to a tube that ran through a soundproof and lightproof lock in the wall to an adjacent room. This allowed us to collect venous blood samples without disturbing subjects' sleep. Blood samples were drawn every 20 minutes, immediately put on ice, and centrifuged within four hours.

Data analyses

Sleep Questionnaires

The Pittsburgh Sleep Quality Index (PSQI) and addendum of the PSQI for PTSD, which were translated into Dutch, were filled out within one month before the sleep registrations (Buysse et al., 1989; Germain et al., 2005). In addition, a sleep log was filled out by the

participants every morning upon awakening during one week at home. We calculated mean total sleep time (MTST), mean sleep onset latency (MSOL), and mean subjective wake after sleep onset (MWASO) for the seven nights in the home situation.

A sleep log assessing subjective sleep onset latency (SOL), total sleep time (TST) and wake after sleep onset (WASO) was also filled out on the morning after the PSG registration in the sleep laboratory. Additionally, participants marked their perception of sleep depth on a scale of 0 to 100 on a visual analogue scale (VAS). Finally, participants were asked to grade habitual sleep (in the last few weeks) and sleep during the recording night with a rating between 0 and 10.

Sleep registrations

Sleep data was analyzed in 30 second epochs according to the criteria of Rechtschaffen and Kales (1968) by an experienced sleep technician who was blind to group identity. Additionally, awakening from S2, SWS and REM sleep, occurring after 2 minutes of continuous sleep, were scored as a measure of sleep fragmentation.

Furthermore, mean heart rate was calculated automatically with the BrainLab software in those without antihypertensive medication (PTSD n=3), or frequent extra systoles (PTSD n=1, HC n=1) (Brain Lab, TMS, the Netherlands). Due to technical problems the PSG of three participants failed (PTSD n=1, TC n=2).

Blood sampling

As mentioned above we adjusted the 8 hour sleep period to the habitual bed times of the participants in order to adjust for differences in circadian rhythm. From now on, we will refer to the normalized times: by definition, “2300h” is time of lights out and “0700h” is time of lights on. Plasma concentrations of cortisol were determined every 20 minutes from 2200h until 0800hr. ACTH levels were determined every hour from 2200h until 0700h. Additionally, ACTH samples were collected at 20 minutes, 40 minutes and 60 minutes after awakenings to determine the awakening curve response (ACR). The ACR is a frequently used and a distinct biological marker for HPA-axis activity (Clow et al., 2010;Pruessner et al., 1997).

Only for melatonin concentrations absolute times were used. Melatonin concentrations were determined hourly from three hours before lights out (between 1900h and 2100h) until awakening (between 0600h and 0800h).

We excluded participants from the cortisol analyses in case of 3 sequentially missed time points (PTSD n = 4, TC = 4, HC = 2), or 2 sequentially missed time points for ACTH (PTSD n = 4, TC = 4, HC = 2) and melatonin (PTSD n = 3, TC = 4, HC = 1). Single missed time points were interpolated, calculating the mean of the two adjoining measured values. All plasma samples were stored at -80 C and analyzed in one batch. ACTH and cortisol were measured using an electrochemiluminescence immunoassay on the Modular E170

(Roche Diagnostics GmbH, Mannheim, Germany). The lower limit of detection of ACTH was 1.0 ng/L and interassay variation was 9 – 5% at 3.5 – 135 ng/L respectively (n = 17). The lower limit of detection of cortisol was 3 nmol/L and interassay variation was 5 – 3% at 290 – 1400 nmol/L respectively (n = 23). Melatonin was measured with a competitive radioimmunoassay (Melatonin Direct RIA KIPL3300,Diasource Immunoassays, Nivelles, Belgium). The lower limit of detection was 2 ng/L. Within-run variation was 10% and 15% at the level of 20 and 60 ng/L respectively. Area under the curve with zero as baseline (AUCg) was computed for cortisol, ACTH, and melatonin (Pruessner et al., 2003). Additionally, AUCg was determined for cortisol and ACTH in the first half of the night, when SWS occurs. The ratio of cortiso:ACTH for the night (2200h – 0700h) and for the ACR was determined as a measure of responsivity of the adrenals upon ACTH stimulation.

Melatonin secretion onset was defined as the time point at which: 1. at least a 50% increase was seen relative to the previous measurement, and 2. the time point was followed by a higher measurement.

Statistical analyses

Differences in demographic variables and objective sleep between groups were tested with ANOVA for three independent samples. Group differences for plasma cortisol, ACTH and melatonin between 2300h and 0700h, and for ACR of cortisol and ACTH were analyzed with a repeated measures ANOVA. BMI was added as a covariate, while BMI tended to be higher in PTSD, and BMI is related to cortisol levels (Rutters et al., 2010). A MANCOVA with BMI as a covariate was used to test for differences when variables were normally distributed (AUCg ACTH, nocturnal ratio cortisol:ACTH, ACR ratio cortisol:ACTH). For variables that were not normally distributed after log transformations, a nonparametric Kruskal-Wallis test for three groups was applied, followed by a Mann-Whitney test for two groups in case of significant differences. P values were Bonferroni corrected for multiple testing.

For dichotomous variables a chi square test was used. Correlations were analyzed with a two-tailed Pearson's test, or with a nonparametric Spearman's rho when variables were not normally distributed after log transformations. Partial correlation analysis, with group as covariate, was performed to test the relationship between ACTH, cortisol, and awakenings. The relationship between SWS and the AUCg during the first half of the night of cortisol and ACTH was also tested with partial correlations, corrected for group. Additional regression analyses were performed for SWS as dependent, with HPA axis parameters and group as regressors. Differences were considered significant when p values were smaller than 0.05.

RESULTS

Demographic data

Table 1 shows demographic characteristics for all participants. PTSD patients and TCs were well matched for deployment and traumatic experiences (table 2). HCs experienced and witnessed some traumatic events, but significantly less than TCs and PTSD patients. Moreover, HCs did not report any complaints related to the reported events in the last month.

Subjective sleep quality

PTSD patients exhibited significantly poorer subjective sleep quality as compared to TCs and HCs according to the PSQI, PSQI addendum and the 7days sleep log (table 3). In addition, PTSD patients had lower scores on subjective sleep measures during the recording night.

Table 1. Demographic and medical characteristics

Demographic characteristics	PTSD (n=13)	TC (n=17)	HC (n= 15)	Group comparison
Age (years) *M (SD)*	37.69 (6.55)	37.18 (6.08)	34.53 (8.18)	F (2,42) =0.87,p = 0.43
Non-Caucasian *n* (%)	1 (8%)	1 (6%)	2 (13%)	χ^2= 0.58, df = 2, p = 0.75
Married *n* (%)	12 (92%)	16 (94%)	5 (33%)	χ^2= 23.89, df = 4, p< 0.001
Currently fulltime work *n* (%)	7 (53%)	16 (94%)	12 (80%)	χ^2= 21.50, df = 10, p= 0.018
Education* *M (SD)*	4.08 (1.38)	5.71 (1.05)	6.13 (1.06)	F (2,42) =12.11,p < 0.001 PTSD < TC, p=0.001 PTSD < HC p<0.001 TC = HC, p=0.91
Medical characteristics				
BMI (kg/m^2) *M (SD)*	26.07 (4.39)	23.84 (2.32)	23.33 (2.40)	F (2,42) =3.01,p = 0.057
Cardiovascular medication *n* (%)	3 (23%)	0	0	χ^2= 7.91, df = 2, p= 0.019
Smoking *n* (%)	4 (31%)	3 (18%)	2 (13%)	χ^2= 1.41, df = 2, p= 0.492
Alcohol (units/week) *M (SD)*	5.92 (7.52)	5.59 (8.17)	7.20 (5.62)	F (2,42) =0.99,p = 0.38
Coffee (cups/day) *M (SD)*	4.15 (3.63)	3.59 (3.20)	2.70 (2.12)	F (2,42) =0.83,p = 0.44
Habitual recreational drugs *n (%)*	0	1 (6%)	3 (20%)	χ^2= 3.75, df = 2, p= 0.15

BMI= Body Mass Index, HC = Healthy Controls, M = Mean, PTSD= Post-traumatic Stress Disorder, SD = Standard Deviation, TC= Trauma Controls

* coded variable (0 = primary school, 7 = university)

Polysomnography

Sleep architecture did not differ between groups (See table 4.) However, PTSD patients had a higher number of awakenings during sleep, 18.67 (5.38) versus 11.47 (4.20) in TCs and 11.07 (3.43) in HCs (F (2,42) = 12.55, p < 0.001). Subjective reports of sleep depth

Table 2. Traumatic events in PTSD and controls.

Psychometric characteristics	PTSD (n=13)	TC (n=17)	HC (n=15)	Group comparison
CAPS score *M (SD)*	64.11 (7.93)	3.65 (4.31)	0	F (2,42) =715.96,p < 0.001 PTSD > TC, p<0.001 PTSD >HC p<0.001 TC = HC, p= 0.14
A 1 trauma of CAPS *M (SD)*	8.92 (2.00)	7.23 (1.72)	0.80 (0.78)	F (2,42) = 52.64, p <0.001 PTSD > TC, p=0.024 PTSD >HC p<0.001 TC >HC p<0.001
HAM D *M (SD)*	14.33 (4.01)	1.27 (1.39)	1.29 (1.44)	F (2,38) =119.31,p < 0.001 PTSD > TC, p<0.001 PTSD >HC p<0.001 TC = HC, p= 1.0
MDD *n* (%)	Current 4 (31%) Lifetime 8 (62%)	Current 0 Lifetime 0	Current 0 Lifetime 0	χ^2= 10.81, df = 2, p=0.005 χ^2= 23.95, df = 2, p< 0.001
Deployment characteristics				
> 1 missions *n (%)*	4 (31%)	6 (35%)	-	χ^2= 0.07, df = 1, p= 0.79
Year, first mission *M (SD)*	1995.00 (5.87)	1994.65 (6.39)	-	U = 103.00, df = 1, p = 0.77
Year, second or last mission *M (SD)*	2003.75 (5.25)	2002.83 (5.19)	-	U = 10.50, df = 1, p = 0.76
Area, first mission *n (%)*	Afghanistan 0 Bosnia 10 Cambodia 0 Iraq 2 Lebanon 1	Afghanistan 1 Bosnia 10 Cambodia 2 Iraq 2 Lebanon 2	-	χ^2= 2.85, df = 4, p= 0.58
Rank, first mission *n (%)*	Soldier 2 Corporal 5 Non-com 2 Officer 1 Missing 2	Soldier 9 Corporal 3 Non-com 1 Officer 2 Missing 2	-	χ^2= 4.81, df = 3, p= 0.19

CAPS= Clinical Administered PTSD Scale, HAMD= Hamilton Depression Scale, HC = Healthy Controls, M = Mean, MDD = Major Depressive Disorder, n = Number, Non-Com = Non-Commissioned officer, PTSD= Post-traumatic Stress Disorder, SD = Standard Deviation, TC= Trauma controls

correlated significantly with the number of awakenings in the group as a whole (r = -.602, p < 0.001, Spearman's rho, n=41). A positive correlation was also observed when only PTSD patients entered the analysis (r = -.616, p = 0.043, n=11) (fig.1).

Heart rate

The mean heart rate of PTSD patients was 64.66 (5.63) beats per minute, and was significantly increased compared with TCs, 57.44 (6.04), but not significantly different from HCs, 58.01 (7.25) (F (2,34) = 3.66, p =0.036. PTSD >TC, p=0.046. PTSD < HC, p=0.078, fig. 2).

Table 3. Subjective sleep quality.

Subjective sleep quality *M (SD)*	**PTSD (n=13)**	**TC (n=17)**	**HC (n= 15)**	**Group comparison**
PSQI	11.54 (3.07)	4.12 (1.69)	3.50 (2.38)	*F (2,41) = 48.14, p<0.001* PTSD > TC, PTSD >HC, p<0.001 TC =HC, p=1.0
Add PSQI	4.92 (2.53)	0.18 (0.53)	0.21 (0.43)	$H^2_{(2)}$ *= 33.68, df =2, p <0.001* PTSD>TC, PTSD>HC, p<0.001 TC = HC, p=0.53
7 days sleep calendar	**(n=11)**	**(n=17)**	**(n= 13)**	
TST (minutes)	362.10 (95.10)	412.65 (40.80)	426.15 (37.05)	$H^2_{(2)}$ *= 5.60, df =2, p=0.061*
WASO (minutes)	68.85 (48.90)	24.00 (20.40)	24.45 (21.30)	*F (2,38) = 8.49, p =0.001* PTSD > TC, p=0.002 PTSD >HC, p=0.003 TC =HC, p=1.0
SOL (minutes)	30.90 (12.15)	15.90 (13.35)	17.25 (10.50)	*F (2,38) = 5.70, p =0.007* PTSD > TC, p=0.009 PTSD >HC, p=0.028 TC =HC, p=1.0
Recording night	**(n=12)**	**(n=17)**	**(n= 15)**	
TST (minutes)	345.60 (92.25)	381.15 (34.35)	394.05 (50.25)	$H^2_{(2)}$ = 2.05, *df* =2, p=0.36
WASO (minutes)	98.70 (59.85)	51.15 (21.90)	52.05 (33.00)	*F (2,41) = 6.40, p=0.004* PTSD > TC, p=0.007 PTSD >HC, p=0.011 TC =HC, p=1.0
SOL (minutes)	33.15 (40.65)	22.05 (16.05)	30.00 (23.40)	$H^2_{(2)}$ = 0.77, *df* =2, p=0.68
No. awakenings	3.58 (1.83)	2.88 (1.17)	3.27 (1.34)	*F (2,41) =0.52, p=0.6*
Sleep efficiency (TST/sleep period time)	72.23 (17.76)	84.4 (5.87)	82.97 (9.98)	$H^2_{(2)}$ = 5.51, *df* =2, p=0.064
VAS sleep dept (0-100)	44.75 (25.79)	75.29 (20.12)	74.80 (10.8)	$H^2_{(2)}$ = 12.46, *df* =2, p=0.002 PTSD < TC, p=0.002 PTSD <HC, p=0.002 TC = HC, p= 0.39
Grade normal night (0-10)	5.83 (0.84)	8.00 (0.69)	7.90 (0.71)	*F (2,41) = 36.18,p = 0.001* PTSD<TC, PTSD<HC, p<0.001 TC =HC p=1.0
Grade recording night (0-10)	5.7 (1.6)	7.12 (0.80)	7.27 (0.93)	*F (2,41) =7.84,p =0.004* PTSD > TC, p=0.005 PTSD >HC p=0.002 TC =HC p=1.0

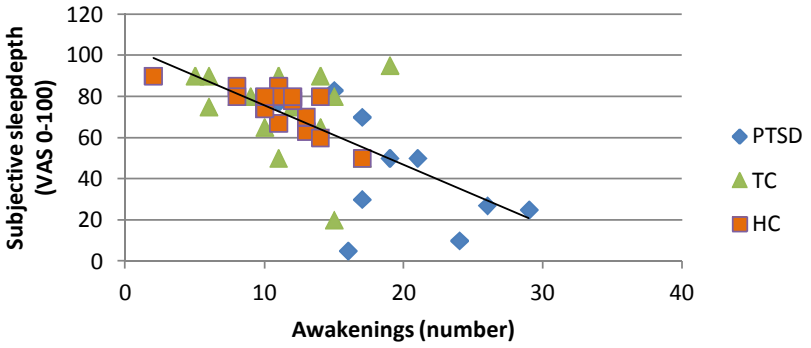
HC = Healthy Controls, M = Mean, PTSD= Posttraumatic Stress Disorder, SD = Standard Deviation, SOL = Sleep Onset Latency, TC= Trauma Controls, TST = Total Sleep Time, VAS = Visual Analogue Scale, WASO = Wake After Sleep Onset

Table 4. Polysomnography.

Sleep architecture *M (SD)*	**PTSD (n=12)**	**TC (n=15)**	**HC (n= 15)**	**Group comparison**
TST (minutes)	405.38 (43.63)	420.80 (36.74)	416.92 (44.43)	$H^2_{(2)} = 1.07, df = 2, p=0.584$
WASO (minutes)	52.71 (35.25)	35.60 (30.27)	44.76 (47.59)	*F (2,39) = 1.65, p=0. 21*
SOL (minutes)	18.29 (15.93)	19.60 (25.46)	14.33 (13.69)	*F (2,39) = 0.59, p=0.56*
Sleep efficiency	85.33 (8.47)	88.47 (7.34)	88.25 (9.84)	$H^2_{(2)} = 2.22, df = 2, p=0.33$
Stage 1 (% of TST)	6.57 (4.18)	5.07 (2.48)	5.66 (2.94)	*F (2,39) = 0.73, p=0.49*
Stage 2 (% of TST)	50.14 (5.69)	49.41 (6.14)	46.46 (8.28)	*F (2,39) = 1.13, p=0.34*
Stage 3 (% of TST)	9.20 (2.54)	7.03 (2.53)	9.96 (9.85)	$H^2_{(2)} = 4.78$, *df* =2, p=0.092
Stage 4 (% of TST)	12.11 (5.11)	12.98 (7.15)	15.56 (7.53)	$H^2_{(2)} = 1.59$, *df* =2, p=0.45
REMS (% of TST)	22.19 (4.71)	25.52 (5.58)	24.98 (3.34)	*F (2,39) = 1.82, p=0.18*
Awakenings during sleep	**(n=12)**	**(n=15)**	**(n= 15)**	
Total	18.67 (5.38)	11.47 (4.20)	11.07 (3.43)	*F (2,42) = 12.55, p<0.001* PTSD > TC, p<0.001 PTSD > HC, p<0.001 TC=HC, p=1.0
Stage 2 (index/ hour S2)	4.17 (1.53)	2.25 (1.34)	2.41 (1.89)	$H^2_{(2)} = 11.00$, *df* =2, p=0.004 PTSD >TC, p=0.003 PTSD >HC, p=0.004 TC = HC, p=0.85
SWS (index/ hour SWS)	1.14 (0.95)	0.32 (0.49)	0.57 (0.80)	*F (2,42) = 4.15, p=0.023* PTSD > TC, p<0.022 PTSD > HC, p<0.15 TC =HC, p=1.0
REMS (index/ hour REMS)	3.95 (2.21)	2.55 (1.94)	2.45 (1.15)	$H^2_{(2)} = 4.78$, *df* =2, p=0.092
23.00-07.00 (mean beats per minute)	64.66 (5.63)	57.44 (6.04)	58.01 (7.25)	*F (2,34) = 3.66, p=0.036* PTSD >TC, p=0.046 PTSD < HC, p=0.078 TC=HC, p=1.0

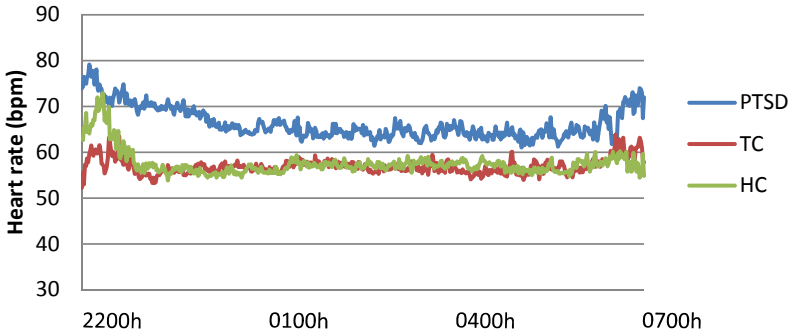
HC = Healthy Controls, M = Mean, PTSD= Post-traumatic Stress Disorder, REMS = Rapid Eye Movement Sleep, SD = Standard Deviation, SOL = Sleep Onset Latency, SWS = Slow Wave Sleep, TC= Trauma Controls, TST = Total Sleep Time, WASO = Wake After Sleep Onset

Figure 1. Relationship between subjective sleep depth and awakenings



r = -0.602, p = 0.000, Spearman's rho, n=41.
HC = Healthy Controls; PTSD = Post-traumatic Stress Disorder, TC = Trauma Controls, VAS = Visual Analogue Scale

Figure 2. Heart rate during sleep



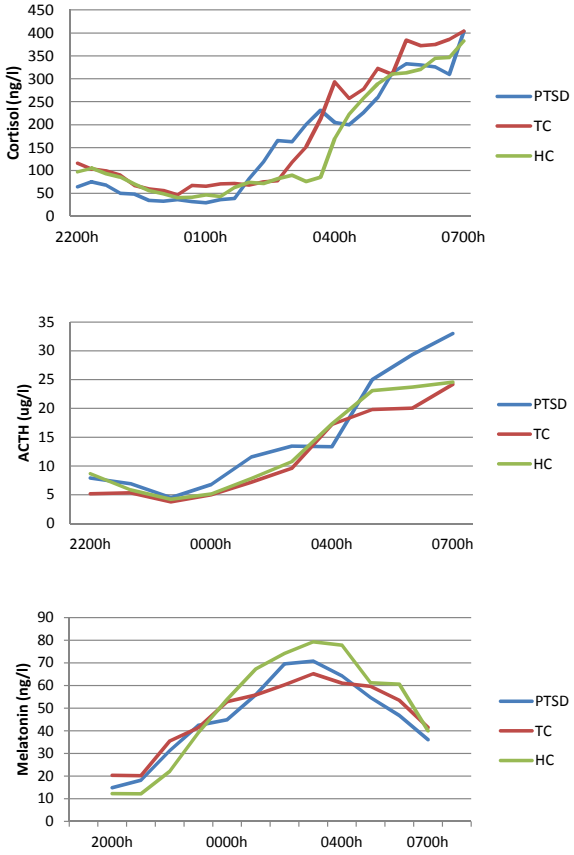
HC = Healthy controls; PTSD = Posttraumatic Stress Disorder, TC = Trauma controls
F (2,34) = 3.66, p =0.036. PTSD >TC, p=0.046. PTSD < HC, p=0.078

Nocturnal plasma cortisol, ACTH, and melatonin concentrations

No differences between the groups in nocturnal cortisol, ACTH or melatonin secretion were found (Fig. 3, table 5). AUCg plasma cortisol between 2200h and 0700h did not differ between groups, although PTSD patients exhibited a trend towards decreased cortisol secretion during the first half of the night (Kruskal-Wallis H_2 =5.75, *df* =2, p = 0.056). No group differences were found for AUCg plasma ACTH and melatonin.

However, the cortisol: ACTH ratio was significantly increased in TCs, 46.16 (11.41) as compared to PTSD patients 31.52 (12.16), and also increased compared to HCs at borderline significance 34.37 (13.01) (F (2,34) = 4.63, P = 0.018, TC > PTSD, p=0.025; TC > HC p = 0.051). Mean times of melatonin onset were similar in the three groups (Table 5).

Figure 3. Nocturnal cortisol, ACTH and melatonin levels



Cortisol: Repeated Measure's ANOVA F (2,36) F =3.13, p = 0.059
ACTH: Repeated Measure's ANOVA F (2,38) = 1.33, p = 0.28
Melatonin: Repeated Measure's ANOVA F (2,34) F =0.36, p = 0.70
HC = Healthy controls; PTSD = Post-traumatic Stress Disorder, TC = Trauma controls

Table 5. Nocturnal levels of cortisol, ACTH and Melatonin.

2300h – 0700h *M (SD)*	**PTSD (n=9)**	**TCs (n=15)**	**HCs (n=13)**	**Group comparison**
Cortisol, AUCg	3748.89 (778.40)	4358.00 (1046.79)	3646.38 (1167.83)	$H^2_{(2)}$ = 3.62, *df* =2, p=0.16
Cortisol 2200h –0120h	540.00 (374.98)	789.86 (401.70)	640.43 (394.18)	$H^2_{(2)}$ = 5.75, *df* =2, p=0.056
ACTH, AUCg	130.56 (47.17)	100.60 (36.87)	113.18 (48.23)	F (2,34) = 0.873, p=0.43
ACTH 2200h –0120h	41.11 (18.75)	28.69 (16.32)	31.79 (20.24)	F (2,34) = 1.88, p=0.17

Table 5. (continued)

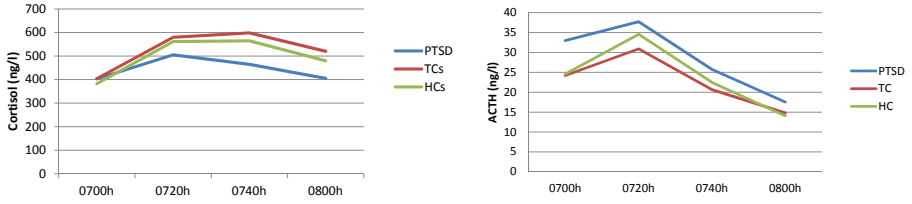
2300h – 0700h *M (SD)*	**PTSD (n=9)**	**TCs (n=15)**	**HCs (n=13)**	**Group comparison**
cortisol:ACTH	31.52 (12.16)	46.16 (11.41)	34.37 (13.01)	F (2,34) = 4.63, p=0.018 TC > PTSD, p=0.025 TC > HC, p=0.051 PTSD = HC, p=1.00
Cortisol:ACTH 2200h –0120h	15.57 (10.79)	36.66 (21.30)	28.23 (24.08)	F (2,34) = 2.88, p=0.07
Melatonin, AUCg	525.56 (410.55)	530.78 (248.98)	636.50 (283.32)	F (2,24) = 0.483, p=0.62
Melatonin onset	2212h (47.50)	2220h (92.40)	2225hr (64.20)	$H^2_{(2)}$ =0.26, *df* =2, p=0.99
Awakening curve response	**(n=10)**	**(n=15)**	**(n=14)**	
Cortisol, AUCg	1423.77 (275.14)	1646.39 (239.70)	1512.11 (308.45)	$H^2_{(2)}$ = 3.18, *df* =2, p=0.204
ACTH, AUCg	93.31 (33.45)	69.75 (27.63)	78.19 (39.83)	$H^2_{(2)}$ = 4.06, *df* =2, p=0.132
cortisol:ACTH (AUCg)	16.86 (5.44)	26.68 (9.70)	22.59 (9.03)	F (2,34) = 4.12, p=0.026 PTSD < TC, p=0.020 PTSD = HC, p=0.322 TC = HC, p=0.649

ACTH = Corticotrophic Hormone, AUCg = Area Under the Curve, HC = Healthy Controls, M = Mean, PTSD = Posttraumatic Stress Disorder, SD = Standard Deviation, TC= Trauma controls

Awakenings Curve Response (ACR)

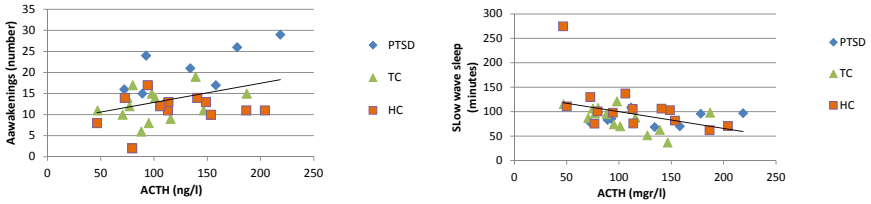
No group differences for cortisol and ACTH levels between 0700h and 0800h were found (Figure 4; table 5). However, the ratio of cortisol: ACTH was significantly decreased in PTSD compared to TC. The ACR was not related to any PSG parameter.

Figure 4. Awakening curve response, cortisol, and ACTH



Cortisol Repeated Measures ANOVA F (2,38) F =1.92, p = 0.162
ACTH Repeated Measures ANOVA F (2,38) F =1.16, p = 0.33
HC = Healthy Controls; PTSD = Post-traumatic Stress Disorder, TC = Trauma Controls.

Figure 5. Relationship between ACTH and objective sleep quality



Awakenings: Pearson's correlation r = .40, df 31, p = 0.022
Slow wave sleep: Pearson's correlation r = -.41, df 31, p = 0.017
HC = Healthy Controls; PTSD = Post-traumatic Stress Disorder, TC = Trauma Controls

Relationship between HPA axis activity and PSG

Partial correlation analyses, controlling for group, showed that AUCg ACTH was positively related to the number of awakenings (r = .40, df 31, p = 0.022), while cortisol was not (r = -.19, df 31, p = 0.29). Furthermore, SWS was inversely related to ACTH secretion (r = -.41, df 31, p = 0.017) and cortisol secretion (r = -.38, df 31, p = 0.028). An additional regression analyses showed that only ACTH was an independent predictor of SWS.

DISCUSSION

This is the first study that measured plasma levels of cortisol, ACTH and melatonin, polysomnographic (PSG) recordings and subjective sleep parameters simultaneously in veterans with PTSD, trauma controls (TCs) and healthy controls (HCs). Heart rate was significantly higher throughout the night in patients with PTSD. They also exhibited more awakenings during sleep according to PSG than the two control groups. ACTH was positively related to the number of awakenings. Furthermore, subjectively perceived sleep quality was related to the number of awakenings on the PSG. ACTH secretion and cortisol were inversely related to the amount of slow wave sleep (SWS). In the first half of the night, a trend was seen for lower levels of cortisol in PTSD. The ratio cortisol: ACTH was decreased upon awakening in PTSD compared with TCs. In TCs the nocturnal cortisol: ACTH ratio was increased compared with both PTSD as TCs.

We hypothesized that the sympathetic nervous system (SNS) would be involved in PTSD related sleep complaints. We did indeed found increased heart rate (HR) in PTSD patients in comparison with TCs and HCs, which indicates increased activity if the SNS, and which is in agreement with previous work (Muraoka et al., 1998; Woodward et al., 2009). However, we failed to find any relationships between HR and the number of awakenings, nor with other sleep disturbances. This may have been due to the small sample size; only 8 patients could enter analyses after excluding those with cardiovascular medication and an irregular cardiac rhythm.

Sleep fragmentation has been previously reported in the absence of major changes in sleep structure and total sleep time in PTSD (Breslau et al., 2004; Germain and Nielsen, 2003; Habukawa et al., 2007; Mellman et al., 1995). The relationship between ACTH secretion and awakenings in PTSD is a novel finding. A relationship between (urinary) cortisol and SWS has also been reported in a previous study (Otte et al., 2005). In our study cortisol and ACTH appeared to be related to SWS. Regression analyses showed that only ACTH was an independent predictor for SWS, in contrast to cortisol. The relationship between ACTH and SWS may be attributed to centrally increased corticotrophic hormone (CRH), which stimulates ACTH secretion form the pituitary. Increased CRH levels have been reported in PSTD in cerebrospinal fluid (Baker et al., 1999) and plasma (de Kloet et al., 2008). CRH has been suggested to inhibit SWS (Neylan et al., 2003; Steiger, 2007). The relationship between awakenings and ACTH may also be due to increased CRH (Neylan et al., 2003; Steiger, 2007).

Interestingly, increased heart rate, which was a robust finding in PTSD, was not related to sleep quality. In contrast, ACTH levels during the night were not significantly altered in PTSD, and were related to SWS and the number of awakenings. It is unknown how these observations should be interpreted. Possibly, we lacked to power to find significant correlation between HR and sleep parameters. Alternatively, increased SNS activity is unrelated to sleep regulation and unrelated to CRH and HPA-axis functioning in PTSD. This has been described in patients with major depressive disorder, in whom the two systems seem to operate autonomously, while in healthy subjects, the HPA-axis and SNS have a reciprocal relationship (Young et al., 2005).

All three groups showed a regular circadian rhythm of cortisol and ACTH secretion. Moreover, there were no group differences in melatonin secretion.

To our knowledge, we are the first to show a relationship between awakenings and subjective sleep quality in PTSD. Only one previous study aimed to assess the relationship between objective sleep measures and subjective reports, for which they used a sleep questionnaire that was administered 1 to 2 months before PSG recordings (Woodward et al., 1996). They did not find a relationship between objective and subjective sleep. However, they did not report the number of awakenings and had no information on the subjective sleep quality of the recording night.

Some group differences in HPA-axis parameters were not related to sleep parameters. Firstly, the awakening curve response (ACR) showed a relative hypocortisolemia in PTSD as opposed to TCs. Secondly, the cortisol: ACTH ratio was increased in TCs as compared to PTSD patients and HCs. A blunted ACR has been reported in PTSD before (de Kloet et al., 2007; Rohleder et al., 2004; Wessa et al., 2006). We also measured ACTH levels, which allowed us to determine the ratio of cortisol: ACTH. This provided information on the cortisol secretion of the adrenals in response to ACTH, suggesting decreased responsiveness of the adrenals in PTSD. An increased ratio of cortisol: ACTH in TCs has previously been

reported (Golier et al., 2007). Hyperresponsive adrenals may possibly reflect an adaptive response to trauma in trauma-exposed individuals without PTSD.

There are several limitations in the interpretation of our findings. First, the small sample size is a limitation of the study. The power to detect differences in cortisol and ACTH levels in the first half of the night, as well as melatonin secretion, was low. Despite the small sample size we were able to find some robust differences between groups. Secondly, some patients used antihypertensive medication. It is well known that beta blockers may increase nightmares complaints, although patients in our study denied experiencing an increase of nightmare frequency after starting medication. Polysomnographic alterations have been described for calcium channel antagonists, but not for other hypertensive agents (Nerbass et al., 2011; Smith et al., 2006). Analyses of awakenings and other PSG data showed similar results when the two patients that used a calcium channel antagonist were excluded from the analyses. Lastly, we did not calculate spectral power of the EEG, which may have enabled us to observe decreased delta activity in PTSD. We found a correlation between SWS and ACTH, while SWS was not decreased in PTSD. Possibly, delta activity would have been decreased, as some previous studies have shown in PTSD. It would have been interesting to examine whether delta activity was related to plasma ACTH concentrations.

In summary, sympathetic activity during sleep is increased in patients with PTSD. Their sleep is fragmented by an increased number of awakenings. Sleep fragmentation is related to HPA axis activity. Future research on the interrelatedness of increased CRH and increased sympathetic activity as neurobiological underpinnings of sleep disturbances in PTSD is warranted. This may help to identify targets for therapeutic interventions for disordered sleep in patients with PTSD.

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REFERENCE LIST

Baker DG, West SA, Nicholson WE, Ekhator NN, Kasckow JW, Hill KK, Bruce AB, Orth DN, Geracioti TD, Jr. (1999). Serial CSF corticotropin-releasing hormone levels and adrenocortical activity in combat veterans with posttraumatic stress disorder. Am J Psychiatry 156:585-588.

Belleville G, Guay S, Marchand A (2009). Impact of sleep disturbances on PTSD symptoms and perceived health. J Nerv Ment Dis 197:126-132.

Blake DD, Weathers FW, Nagy LM, Kaloupek DG, Gusman FD, Charney DS, Keane TM (1995). The development of a Clinician-Administered PTSD Scale. J Trauma Stress 8:75-90.

Breslau N, Roth T, Burduvali E, Kapke A, Schultz L, Roehrs T (2004). Sleep in lifetime posttraumatic stress disorder: a community-based polysomnographic study. Arch Gen Psychiatry 61:508-516.

Buysse DJ, Reynolds CF, III, Monk TH, Berman SR, Kupfer DJ (1989). The Pittsburgh Sleep Quality Index: a new instrument for psychiatric practice and research. Psychiatry Res 28:193-213.

Clow A, Hucklebridge F, Thorn L (2010). The cortisol awakening response in context. Int Rev Neurobiol 93:153-175.

Davidson JR, Rothbaum BO, van der Kolk BA, Sikes CR, Farfel GM (2001.) Multicenter, double-blind comparison of sertraline and placebo in the treatment of posttraumatic stress disorder. Arch Gen Psychiatry 58:485-492.

de Kloet CS, Vermetten E, Geuze E, Lentjes EG, Heijnen CJ, Stalla GK, Westenberg HG (2008.) Elevated plasma corticotrophin-releasing hormone levels in veterans with posttraumatic stress disorder. Prog Brain Res 167:287-291.

de Kloet CS, Vermetten E, Heijnen CJ, Geuze E, Lentjes EG, Westenberg HG (2007). Enhanced cortisol suppression in response to dexamethasone administration in traumatized veterans with and without posttraumatic stress disorder. Psychoneuroendocrinology 32:215-226.

Germain A, Hall M, Krakow B, Katherine SM, Buysse DJ (2005). A brief sleep scale for Posttraumatic Stress Disorder: Pittsburgh Sleep Quality Index Addendum for PTSD. J Anxiety Disord 19:233-244.

Germain A, Nielsen TA (2003). Sleep pathophysiology in posttraumatic stress disorder and idiopathic nightmare sufferers. Biol Psychiatry 54:1092-1098.

Golier JA, Schmeidler J, Legge J, Yehuda R (2007). Twenty-four hour plasma cortisol and adrenocorticotropic hormone in Gulf War veterans: relationships to posttraumatic stress disorder and health symptoms. Biol Psychiatry 62:1175-1178.

Habukawa M, Uchimura N, Maeda M, Kotorii N, Maeda H (2007). Sleep findings in young adult patients with posttraumatic stress disorder. Biol Psychiatry 62:1179-1182.

Hurwitz TD, Mahowald MW, Kuskowski M, Engdahl BE (1998). Polysomnographic sleep is not clinically impaired in Vietnam combat veterans with chronic posttraumatic stress disorder. Biol Psychiatry 44:1066-1073.

Kim E, Grover LM, Bertolotti D, Green TL (2010). Growth hormone rescues hippocampal synaptic function after sleep deprivation. Am J Physiol Regul Integr Comp Physiol 298:R1588-R1596.

Klaassens ER, Giltay EJ, van VT, Veen G, Zitman FG (2010). Trauma exposure in relation to basal salivary cortisol and the hormone response to the dexamethasone/CRH test in male railway employees without lifetime psychopathology. Psychoneuroendocrinology 35:878-886.

Kobayashi I, Boarts JM, Delahanty DL (2007). Polysomnographically measured sleep abnormalities in PTSD: a meta-analytic review. Psychophysiology 44:660-669.

Mellman TA, Kulick-Bell R, Ashlock LE, Nolan B (1995). Sleep events among veterans with combat-related posttraumatic stress disorder. Am J Psychiatry 152:110-115.

Muraoka MY, Carlson JG, Chemtob CM (1998). Twenty-four-hour ambulatory blood pressure and heart rate monitoring in combat-related posttraumatic stress disorder. J Trauma Stress 11:473-484.

Nerbass FB, Pedrosa RP, Genta PR, Drager LF, Lorenzi-Filho G (2011). Calcium channel blockers are independently associated with short sleep duration in hypertensive patients with obstructive sleep apnea. J Hypertens 29:1236-1241.

Neylan TC, Lenoci M, Maglione ML, Rosenlicht NZ, Metzler TJ, Otte C, Schoenfeld FB, Yehuda R, Marmar CR (2003). Delta sleep response to metyrapone in post-traumatic stress disorder. Neuropsychopharmacology 28:1666-1676.

Neylan TC, Marmar CR, Metzler TJ, Weiss DS, Zatzick DF, Delucchi KL, Wu RM, Schoenfeld FB (1998). Sleep disturbances in the Vietnam generation: findings from a nationally representative sample of male Vietnam veterans. Am J Psychiatry 155:929-933.

Neylan TC, Mueller SG, Wang Z, Metzler TJ, Lenoci M, Truran D, Marmar CR, Weiner MW, Schuff N (2010). Insomnia severity is associated with a decreased volume of the CA3/dentate gyrus hippocampal subfield. Biol Psychiatry 68:494-496.

Nielsen T (2010). Nightmares associated with the eveningness chronotype. J Biol Rhythms 25:53-62.

Ohayon MM, Shapiro CM (2000). Sleep disturbances and psychiatric disorders associated with post-traumatic stress disorder in the general population. Compr Psychiatry 41:469-478.

Otte C, Lenoci M, Metzler T, Yehuda R, Marmar CR, Neylan TC (2005). Hypothalamic-pituitary-adrenal axis activity and sleep in posttraumatic stress disorder. Neuropsychopharmacology 30:1173-1180.

Pillar G, Malhotra A, Lavie P (2000). Post-traumatic stress disorder and sleep-what a nightmare! Sleep Med Rev 4:183-200.

Pruessner JC, Kirschbaum C, Meinlschmid G, Hellhammer DH (2003). Two formulas for computation of the area under the curve represent measures of total hormone concentration versus time-dependent change. Psychoneuroendocrinology 28:916-931.

Pruessner JC, Wolf OT, Hellhammer DH, Buske-Kirschbaum A, von AK, Jobst S, Kaspers F, Kirschbaum C (1997). Free cortisol levels after awakening: a reliable biological marker for the assessment of adrenocortical activity. Life Sci 61:2539-2549.

Rechtschaffen A, Kales A (1968). A manual of Standardized Terminology, Techniques and Scoring System for Sleep Stages of Human Subjects. Washington DC: US Government Printing Office.

Ribeiro JD, Pease JL, Gutierrez PM, Silva C, Bernert RA, Rudd MD, Joiner TE, Jr. (2011). Sleep problems outperform depression and hopelessness as cross-sectional and longitudinal predictors of suicidal ideation and behavior in young adults in the military. J Affect Disord.

Rohleder N, Joksimovic L, Wolf JM, Kirschbaum C (2004). Hypocortisolism and increased glucocorticoid sensitivity of pro-Inflammatory cytokine production in Bosnian war refugees with posttraumatic stress disorder. Biol Psychiatry 55:745-751.

Rutters F, Nieuwenhuizen AG, Lemmens SG, Born JM, Westerterp-Plantenga MS (2010). Hypothalamic-pituitary-adrenal (HPA) axis functioning in relation to body fat distribution. Clin Endocrinol (Oxf) 72:738-743.

Saper CB, Scammell TE, Lu J (2005). Hypothalamic regulation of sleep and circadian rhythms. Nature 437:1257-1263.

Smith SS, Dingwall K, Jorgenson G, Douglas J (2006). Associations between the use of common medications and sleep architecture in patients with untreated obstructive sleep apnea. J Clin Sleep Med 2:156-162.

Spitzer RL, Williams JB, Gibbon M, First MB (1992). The Structured Clinical Interview for DSM-III-R (SCID). I: History, rationale, and description. Arch Gen Psychiatry 49:624-629.

Steiger A (2007). Neurochemical regulation of sleep. J Psychiatr Res 41:537-552.

Van Der Werf YD, Altena E, Schoonheim MM, Sanz-Arigita EJ, Vis JC, De RW, Van Someren EJ (2009). Sleep benefits subsequent hippocampal functioning. Nat Neurosci 12:122-123.

van Liempt S, Vermetten E, Lentjes E, Arends J, Westenberg H (2011). Decreased nocturnal growth hormone secretion and sleep fragmentation in combat-related posttraumatic stress disorder; potential predictors of impaired memory consolidation. Psychoneuroendocrinology 36:1361-1369.

Wessa M, Rohleder N, Kirschbaum C, Flor H (2006). Altered cortisol awakening response in post-traumatic stress disorder. Psychoneuroendocrinology 31:209-215.

Woodward SH, Arsenault NJ, Voelker K, Nguyen T, Lynch J, Skultety K, Mozer E, Leskin GA, Sheikh JI (2009). Autonomic activation during sleep in posttraumatic stress disorder and panic: a mattress actigraphic study. Biol Psychiatry 66:41-46.

Woodward SH, Bliwise DL, Friedman MJ, Gusman DF (1996). Subjective versus objective sleep in Vietnam combat veterans hospitalized for PTSD. J Trauma Stress 9:137-143.

Young EA, Abelson JL, Cameron OG (2005a). Interaction of brain noradrenergic system and the hypothalamic-pituitary-adrenal (HPA) axis in man. Psychoneuroendocrinology 30:807-814.

Zayfert C, DeViva JC (2004). Residual insomnia following cognitive behavioral therapy for PTSD. J Trauma Stress 17:69-73.

Chapter 5

Decreased nocturnal growth hormone secretion and sleep fragmentation in combat-related post-traumatic stress disorder; potential predictors of impaired memory consolidation

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ABSTRACT

Background. Healthy sleep facilitates the consolidation of newly acquired memories. Although patients with post-traumatic stress disorder (PTSD) often complain of sleep disturbances and memory deficits, the interrelatedness of these symptoms is not well understood. Sleep may be disturbed in PTSD by increased awakenings during sleep, which has been associated with decreased growth hormone (GH) secretion. We conducted a controlled study in which we assessed sleep fragmentation, nocturnal secretion of GH, and memory consolidation in patients with PTSD.

Methods. While sleep EEG was being monitored, 13 veterans with PTSD, 15 trauma controls (TC) and 15 healthy controls (HC) slept with an iv catheter, through which blood was collected every 20 minutes from 2300h pm to 0800h. Declarative memory encoding was assessed with the 15 word task before sleep, and consolidation was assessed the next morning by a free recall.

Results. Sleep was more fragmented in patients with PTSD, with more awakenings in the first half of the night ($p < 0.05$). Plasma levels of GH during the night were significantly decreased in PTSD compared with HC ($p < 0.05$). Furthermore, GH secretion and awakenings were independent predictors for delayed recall, which was lower in PTSD compared to HC ($p< 0.05$).

Conclusions. These data show that PTSD is associated with increased awakenings during sleep and decreased nocturnal GH secretion. Furthermore, decreased GH secretion may be related to sleep fragmentation and both variables may exert a negative effect on sleep dependent memory consolidation.

INTRODUCTION

In the last few years evidence has accumulated for the importance of sleep on memory formation. Overnight sleep or a daytime nap strengthens newly formed memory (Stickgold, 2005; Gais et al., 2006; Rasch et al., 2007; Lahl et al., 2008). During sleep hippocampal neurons reactivate and newly acquired information is reorganized and transferred to neocorticol areas for permanent storage (Gais et al., 2007; Takashima et al., 2009). In addition, the ability of the hippocampus to store new information is dependent on previous sleep quality; functional MRI studies have shown that interrupted sleep and sleep deprivation lead to decreased encoding and decreased hippocampus activation during encoding (Yoo et al., 2007; Van Der Werf et al., 2009). Growth hormone (GH), which stimulates neuronal survival and growth, has a peak during the first half of the night (Van Cauter et al. 2004) - a period of enhanced declarative memory consolidation (Rasch et al., 2007). GH receptors are present in the hippocampus (Lai et al., 1993), a brain region implicated in memory consolidation and brain plasticity. A relationship between GH secretion and sleep dependent memory formation has been suggested in a preclinical study (Kim et al., 2010). Some disorders that are accompanied by disturbed sleep, such a Obstructive Sleep Apnea Syndrome (OSAS), and Major Depressive Disorder (MDD), are associated with both impaired cognitive functioning, sleep fragmentation and a blunted GH peaks during the night (Steiger, 2007; Lanfranco et al., 2010).

Amongst the hallmark symptomes of post-traumatic stress disorder are insomnia and nightmares. In addition, PTSD patients often suffer from memory complaints that can affect normal functioning in work, social situations and at home (Geuze et al., 2009; Belleville et al., 2009). Typically, patients remain alert at night and re-experience their traumatic memories during nightmares. Yet, despite sometimes severe sleep complaints, polysomnographic studies have reported only mild disturbances in sleep structure, for a meta analyses see (Kobayashi et al., 2007). However, other studies found evidence for fragmented sleep with an increased number of nocturnal awakenings and arousals (Mellman et al., 1995; Germain and Nielsen, 2003; Habukawa et al., 2007).

To date, nocturnal GH secretion and overnight memory consolidation has not been studied in PTSD. We hypothesize that in PTSD patients sleep fragmentation is related to decreased GH secretion. Furthermore, we hypothesize that decreased GH secretion is associated with poor memory performance.

MATERIALS AND METHODS

Participants

Veterans with PTSD were recruited through the outpatient clinic of the Military Mental Healthcare, Utrecht, the Netherlands. Trauma controls (TC; veterans without PTSD) and healthy controls (HC; service members never deployed or civilians) were recruited through advertisements. Controls were matched with the PTSD group for age, year of deployment (TC) and region of deployment (TC). After a verbal and written description of the study, written informed consent was obtained. All participants were screened for psychiatric illness using the Structured Clinical Interview for DSM IV axis I disorders (SCID-I) (Spitzer et al., 1992). The diagnosis of PTSD was confirmed by the Clinician Administered PTSD Scale (CAPS) (Blake et al., 1995). PTSD patients were included when they had a CAPS score over 50 and did not meet DSM IV criteria for psychiatric disorders other than mood and anxiety disorder (according to the SCID-I). TCs were included when they met the A1 criteria for PTSD (the person experienced, witnessed or was confronted with an event or events that involved actual or threatened death or serious injury, or a threat to physical integrity of self or others), but had a CAPS score below 18 and did not meet DSM-IV criteria for any current or life-time DSM IV axis I disorder. Typically, TC and PTSD patients were deployed to Afghanistan, Bosnia or Lebanon for a period of 4 to 6 months. During deployment they were exposed to war zone stressors as such as enemy fire, armed combat, and seeing seriously injured fellow soldiers and civilians.

HCs were screened for A1 criteria and were excluded when they experienced or witnessed life-threatening events. All participants were medically healthy male and were free from psychotropic medication and alcohol or drugs dependence in the past six months. All control subjects were without a history of psychiatric disorders, and without sleep complaints. While obstructive sleep apnea syndrome (OSAS) and periodic limb movement disorder (PLMD) are accompanied by frequent arousals and awakenings during sleep, all participants who screened positive for OSAS and PLMD with one night of polysomnography (PSG), were excluded from the study. When PSG registrations ruled out OSAS and PLMD, participants were scheduled for 2 nights PSG with simultaneous blood sampling and memory testing.

The study was approved by the Institutional Review Board of the University Medical Centre of Utrecht, The Netherlands.

Procedures

Screening OSAS and PLMD

During one night PSG recordings were made with Brainlab Real Time ©, including EOG for vertical and horizontal eye movements, EMG (chin, left and right m. tibialis anterior),

ECG and EEG (F0-CO, F3-C3, P3-O1, C4-A1, O2-A2), airflow at the nose (nasal pressure), movement of the thorax and abdomen (by inductance plethysmography), and arterial oxygen saturation at the index finger. Apneas and hypopneas were scored when an airflow reduction of 50-90 % (hypopnea) or >90 % (apnea) was detected for at least 10 seconds, and were deleted when occurring after arousals or during wake. OSAS was defined as more than 15 apnea or hypopnea per hour. PLMD was defined as more than five PLM related arousals per hour.

Sleep registrations with simultaneous blood sampling

Sleep registrations during two consecutive nights were conducted in the sleep laboratory of the Central Military Hospital in Utrecht, the Netherlands. Ambulatory sleep recordings were acquired with Twente Medical Systems, Porti 7 (TMS International BV), including bipolar derivations of EMG (chin), EOG for vertical and horizontal eye movements, EEG (F0-CO, F3-C3, P3-O1, C4-A1, O2-A2) and ECG.

The first night served as adaptation to the laboratory setting. We adjusted the 8 hour sleep period to the habitual times of participants, with a time of lights out between 2200h and 0000h, and lights on between 0600h and 0800h. From now on, we will only refer to the normalized times: by definition, "2300h" is time of lights out and "0700h" is time of lights on. Participants slept with an IV catheter in a fore arm vein, which was connected to a tube that ran through a sound and light proof lock in the wall to an adjacent room. This allowed us to collect venous blood samples without disturbing subjects' sleep. Blood samples were drawn every 20 minutes, immediately put on ice, and centrifuged within four hours. Participants were instructed to refrain from alcohol on the day prior to sleep registration, and to refrain from caffeine containing beverages after 1600h on the day prior to sleep registration.

Memory task

To assess declarative memory consolidation the 15 word task (15WT) (Saan and Deelman, 1986) was administered three hours before sleep on the second evening. Fifteen neutral one syllable words were visually presented on a computer screen, and repeated three times. Every presentation was followed by a free immediate recall. A free delayed recall was assessed the next morning between 30 – 45 minutes after awakening. Patients were unaware of the delayed recall task the next morning to prevent rehearsal.

Data analyses

Sleep registration

Sleep data were analyzed in 30 second epochs according to criteria of Rechtschaffen and Kales (1968), by an experienced sleep technician who was blind to PTSD diagnosis. The

number of spontaneous awakenings from Stage 2 (S2) sleep, slow wave sleep (SWS) or rapid eye movement (REM) sleep was determined for the first half of the night. Furthermore, we determined total sleep time (TST), the absolute amount of SWS in minutes, and SWS as a percentage of TST.

Blood sampling

Plasma samples were stored at -80 C and analyzed in one batch. Human Growth Hormone was measured using an immunometric technique on an IMMULITE 1000 Analyzer (Siemens Medical Solutions Diagnostics, Los Angeles, USA). The lower limit of detection was 0.12 mE/L and inter-assay variation was 5,5% at 0.45 mE/L, and 3,2 % at 23 mE/l (n = 23).

Area under the curve (AUC) with zero as baseline (AUCg) and AUC with baseline prior to the increase (AUCi) were calculated (Pruessner et al., 2003). AUCg reflects the total amount of secreted GH during the night. AUCi reflects the amount of GH secreted during the peak.

Statistical analyses

Differences in demographic variables and objective sleep between groups were tested with ANOVA for three independent samples. Group differences of GH secretion between 2320h and 0240h were analyzed with a repeated measures ANOVA with BMI as a covariate; to this end, single missing values were interpolated as a mean value between two measured points. Missing values were only interpolated when a clear peak was present. Subsequently, ANCOVA with BMI as a covariate was performed for each time point and Bonferroni corrected for multiple comparisons. For the analyses of AUCi and AUCg, ANCOVA was performed with BMI as a covariate and a Bonferroni post hoc test to compare differences between the three groups. For dichotomous variables a chi square test was used. We performed a linear regression analysis with delayed recall as a dependent variable and AUCg GH, awakenings, third trial score of the immediate recall and group as regressors. Furthermore, we tested the corrrelation between AUCg GH and awakenings with a Spearman's rho's test, because awakenings were not normally distributed after log transformation. A nonparametric Kruskal Wallis test for group differences was used for variables that were not normally distributed after log transformation. Differences were considered significant when p values were smaller than 0.05.

RESULTS

From the 22 PTSD patients who were recruited to participate, one was excluded because of cannabis dependence, one was lost for follow up, one was excluded due to a BMI of

40, one reported symptom relieve after psychotherapy before the study was finished, one patient was excluded due to OSAS, and three patients screened positive for PLMD. A total of 14 PTSD were included. Three patients used medication for hypertension; beta-blocker (*n*=3), Calcium channel antagonist (*n*=2), ACE inhibitor (*n*=1), but were not excluded from the study. From the 70 TCs who registered to participate, 24 subjects were matched with the patients and participated in the study. Four were excluded because they were screened positive for OSAS. Two TC could not participate due to PLMD, and one TC was no longer available. Seventeen TCs entered the study.

From the 18 HCs who decided to participate after a telephone screening, one was excluded due to severe A1 traumatic events and two HCs could not participate due to OSAS. Fifteen HCs participated in this study.

In one PTSD patient and two TCs sleep recordings failed due to technical problems. In one patient time in bed was only five hours, his data were excluded. The final data set of eight hours PSG recordings consisted of data from 12 PTSD patients, 15 TCs and 15 HCs.

Participants were excluded for GH analyses when three or more consecutive values in the first half of the night were missing (4 PTSD patients, 3 TCs and 3 HCs). Furthermore, outliers with GH peaks higher than 36, based on the mean of all participants (10.8 mE/l) + 3 times the SD (8.45) were excluded (1 PTSD patient, GH peak 39 mE/l).

The final data set for GH analysis consisted of data from 8 PTSD patients, 14 TCs and 12 HCs. Correlations between delayed recall and GH secretion were based on 8 PTSD patients, 14 TCs and 12 HCs.

Furthermore, while the sleep recordings of one patient and two TCs in the GH analyses failed, correlations between sleep fragmentation and GH secretion were based on 7 PTSD patients, 12 TCs and 12 HCs.

Demographic and psychometric variables

Demographic, medical and psychometric characteristics of all participating subjects (13 PTSD patients, 17 TCs and 15 HCs) are displayed in table 1. No group differences were present in age, daily coffee intake and weekly alcohol intake. There were no differences between TCs and PTSD patients in number of deployments and year of deployment. HCs experienced or witnessed some potentially traumatic events, but significantly less than TCs and PTSD patients ($p < 0.001$). Moreover, HCs did not report any complaints in the last month related to the reported events.

Growth Hormone secretion

Repeated measures ANOVA with BMI as a covariate showed a significant GH group interaction (Hotelling's Trace $p = 0.025$) and a significant group effect ($p=0.036$). BMI was not a significant covariate in this analysis ($p = 0.184$). Post hoc analyses showed a significant difference between PTSD patients and HC at 0000h, 0020h, 0040h (ANCOVA,

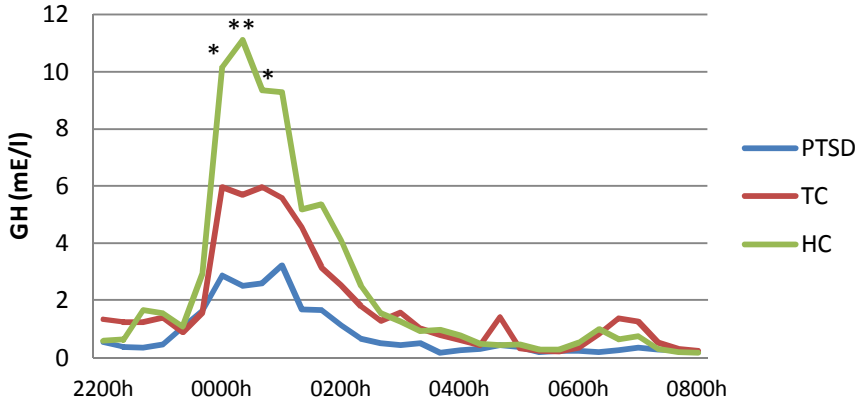
Table 1. Demographic, medical and psychometric characteristics

	PTSD (n=13)	TC (n=17)	HC (n= 15)	Group Comparison
Sociodemographic characteristics				
Age (years)*M (SD)*	37.69 (6.55)	37.18 (6.08)	34.53 (8.18)	p = 0.43
Non-Caucasian *n* (%)	1 (8%)	1 (6%)	2 (13%)	p = 0.75
Married *n* (%)	12 (92%)	16 (94%)	5 (33%)	p < 0.01
Currently fulltime work *n* (%)	7 (53%)	16 (94%)	12 (80%)	p = 0.02
Education* *M (SD)*	4.08 (1.38)	5.71 (1.05)	6.13 (1.06)	p < 0.01
Smoke, yes *n (%)*	4 (31%)	3 (17 %)	2 (13%)	p = 0.49
Alcohol (units/week) *M (SD)*	5.92 (7.52)	5.59 (8.17)	7.20 (5.62)	p = 0.38
Coffee (cups/day) *M (SD)*	4.15 (3.63)	3.59 (3.20)	2.70 (2.12)	p = 0.44
Deployment characteristics				
> 1 missions *n (%)*	4 (31%)	6 (35%)		p = 0.79
Year, first mission *M (SD)*	1995.00 (5.87)	1994.65 (6.39)		p = 0.77
Medical characteristics				
BMI (kg/m^2) *M (SD)*	26.07 (4.39)	23.84 (2.32)	23.33 (2.40)	p = 0.06
Cardiovascular medication *n* (%)	3 (23%)	0	0	p = 0.02
Incidental recreational drugs *n (%)*	0	1 (6%)	3 (20%)	p = 0.09
AHI *M (SD)*	4.8 (3.6)	4.3 (2.9)	4.2 (3.7)	p = 0.79
Psychometric characteristics				
CAPS score *M (SD)*	64.11 (7.93)	3.65 (4.31)	0	p < 0.01
Major Depressive Disorder				
- Current *n* (%)	4 (31%)	0	0	p < 0.01
- Lifetime *n* (%)	8 (62%)	0	0	p < 0.01

AHI = Apnea Hypopnea Index, BMI= Body Mass Index, CAPS= Clinical Administered PTSD Scale, HC = Healthy controls, PTSD = Post-traumatic Stress Disorder, TC = Trauma Controls
* coded variable (0 = primary school, 7 = university)

Bonferroni corrected, resp p = 0.031, p < 0.001 and p = 0.011). GH levels from TCs and PTSD patients were not different on any time point. TCs had a significantly lower GH level than HCs at 0020h (p=0.011). See figure 1.

Figure 1. Nocturnal plasma growth hormone concentration



PTSD (n=8), TC= trauma controls (n=14) , HC = healthy controls (n=12), GH = Growth Hormone
* *PTSD < HC, Repeated measures ANOVA p <0.05*
** *PTSD < HC and TC < HC, Repeated measures ANOVA, p <0.05*

Group differences were observed for both AUCg (ANCOVA p = 0.022), and AUCi (ANCOVA p = 0.014) when corrected for BMI, which was a significant covariate (p =0.013) in this analysis, and corrected for caffeine, which was not a significant covariate (p=0.58) (See table 2). AUCg in PTSD patients (25.6 ± 12.8) was significantly lower than in HCs (76.0 ± 44.8), but not compared with TCs (55.0 ± 43.7) (ANCOVA, Bonferroni corrected, respectively p=0.019, p= 0.210). Similarly, AUCi in PTSD patients (19.1 ± 12.9) was significantly lower than in HCs (58.6 ± 24.2), and unchanged in comparison with TCs (37.1 ± 22.3) (ANCOVA, Bonferroni corrected, resp. p = 0.017, and p= 0.713). There were no significant differences between TCs and HCs in AUCg (ANCOVA, Bonferroni corrected, p = 0.432). A trend was observed for a lower AUCi in TCs as compared with HCs (ANCOVA, Bonferroni corrected, p = 0.079).

Sleep fragmentation and Slow wave sleep

The mean number of awakenings during the first half of the night was significantly higher in PTSD patients (8.5 ± 3.8), than in TCS (4.3 ± 2.8) and HCs (5.1 ± 2.2) (Kruskal-Wallis, respectively p=0.006, and p=0.011, see table 2). TST was 405,4 ± 43,6 in PTSD patients, 420,8 ± 36,7 in TCs and 416,9 ± 44,4 in HCs (Kruskal Wallis, p = 0.532). Similarly, SWS percentage was not decreased in PTSD patients (21.3 ± 5.9) compared with TCs (20.0 ± 5.6) and HCs (25.5 ± 12.6) (ANOVA, p=0.267). Additionally, the absolute amount of SWS (in minutes) was similar in PTSD patients (86.0 ± 17.5) as compared with TCs (83.5 ± 25.17) and HC (92.6 ± 26.0) (ANOVA, p= 0.478).

The number of awakenings showed a negative correlation with AUCg when all subjects entered the analysis (Spearman r = - 0.547, p = 0.004).

Table 2. Growth hormone secretion, sleep and memory in PTSD and controls

Growth Hormone Secretion *M (SD)*	**PTSD (n=8)**	**TC (n=14)**	**HC (n= 12)**	**Group Comparison**
AUCi	19.1 (12.9)	37.1 (22.3)	58.6 (24.2)	P = 0.01[a]
AUCg	25.6 (12.8)	55.0 (43.7)	76.0 (44.8)	p = 0.02[a]
Polysomnography	**(n=12)**	**(n=15)**	**(n= 15)**	
Awakenings 2300h -0300h	8.5 (3.8)	4.3 (2.8)	5.1(2.2)	p = 0.01[b]
% SWS	21.3 (5.9)	20.0 (5.6)	25.5 (12.6)	p = 0.63
Total sleep time	405,4 (43,6)	420,8 (36,7)	416,9(44,4)	p = 0.58
Minutes of SWS	86.0 (17.5)	83.5 (25.17)	92.6 (26.0)	p = 0.48
15 Word Task	**(n=13)**	**(n=15)**	**(n= 15)**	
Third immediate recall	10.8 (2.0)	11.9 (1.7)	12.1 (1.1)	p = 0.62
Post sleep delayed recall	7.5 (2.9)	9.5 (2.5)	10.2 (2.7)	p = 0.03[c]
Correlations				
Awakenings and AUCg	**(n=7)**	**(n=12)**	**(n= 12)**	**All subjects (n=31)**
	r = - 0.55, p = 0.10	r = - 0.33, p = 0.15	r = - 0.56, p = 0.03	r = -0.47, p < 0.01[d]
AUCg and delayed recall	**(n=8)**	**(n=14)**	**(n= 12)**	**All subjects (n=34)**
	r = 0.59, p = 0.06	r = 0.41, p = 0.07	r = 0.58, p = 0.02	r = 0.55, p < 0.01[e]

AUCg = Area under the curve with respect to the ground, AUCi = Area under the curve with respect to the increase, BMI = Body Mass Index, HC = Healthy controls, PTSD = Post-traumatic Stress Disorder, TC = Trauma controls, SWS = slow wave sleep

[a] ANCOVA for three groups with BMI as covariate, PTSD < HC, p < 0.05 (Bonferroni corrected)

[b] Kruskal Wallis test for three groups. PTSD > TC, p = 0.006. PTSD > HC, p = 0.011

[c] ANOVA for three groups, post hoc PTSD < HC, p < 0.05 (Bonferroni corrected)

[d] Spearman's rho correlation

[e] Pearson's correlation

This correlation was also significant for HCs (r = -0.571, p =.033). In PTSD patients and TCs the correlation was not statistically significant (respectively, r = -0.546, p = 0.103, and r = -0.332, p = 0.146).

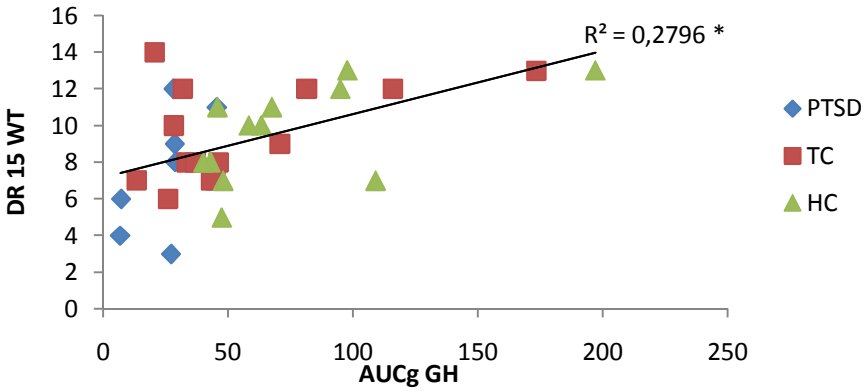
In order to further explore whether caffeine intake (or withdrawal) attributed to the group differences, we performed an ANCOVA with awakenings as depended, group as fixed factor, and caffeine as covariate. Caffeine was not a significant covariate (p=0.31).

Memory task

No group differences were seen in the third immediate recall preceding sleep (ANOVA p = 0.162), yet PTSD patients had a significantly lower score on delayed recall (7.5 ± 2.9) compared to HCs (10.2 ± 2.7) (ANOVA p=0.029, Bonferroni corrected, p= 0.031, see table 2). The mean score in TCs (9.5 ± 2.5) did not differ significantly from either PTSD patients or HCs (Bonferroni respectively p = 0.129, p = 1.00). When performing a repeated measures ANOVA between groups with IR and DR as time points, time was a significant variable p < 0.001, and group*time was not (p=0.191), which indicates that in all three groups a significant decline was present. Also, group was significant (p = 0.04, PTSD < HC p=0.047, bonferroni corrected). The decline was on average larger in PTSD (PTSD 3,3 ± 2,3 TC 2,4 ± 2,0 HC1,9 ± 1,7), but not statistically significant.

Multiple regression analyses showed that awakenings (p=0.024) and GH AUCg (P=0.013) were independent predictors for delayed recall.

Figure 2. Association between growth hormone secretion and delayed recall



AUCg = area under the curve with respect to the ground, DR 15 WT = delayed recall of the 15 word task; GH = growth hormone, PTSD = post traumatic stress disorder (n=8), TC = trauma controls (n=14), HC = healthy controls (n=12).
* Pearson's correlation, r = 0.55, p < 0.01

Figure 2 shows the correlation between delayed recall of the 15WT with the AUCg in all subjects (Pearson's r = 0.55, p = 0.001, see figure 2). This correlation just failed to reach statistical significance in PTSD patients (r = 0.59, p = 0.063) and TCs (r = 0.41, p = 0.074) separately, but remained statistically significant in HCs (r = 0.58, p = 0.023). We repeated the analyses without the two data points with very high GH values and found similar, and significant results (Pearson's test r= 0.428, p=0.007).

DISCUSSION

This is the first study to report on decreased nocturnal growth hormone (GH) secretion in PTSD. Furthermore, a correlation between GH secretion and sleep fragmentation was observed, while total sleep time (TST) and slow wave sleep (SWS) were not altered in PTSD patients. It has been suggested that sleep quality decreased nocturnal GH secretion may negatively affect cognitive functioning. To this end, we performed a declarative memory task before sleep with a post sleep delayed recall and found that both sleep fragmentation and GH secretion predicted performance on the delayed recall task.

TCs exhibited intermediate results on GH secretion and memory performance, which did not statistically differ from both PTSD patients and HCs, with an exception of a lower plasma GH level than HCs at 00.20 h. Possibly, decreased GH secretion in TCs and PTSD patients is the result of alterations in the neurobiology of subjects exposed to chronic stress and psycho-trauma, independently of current psychopathology, which is also the case for alterations in the hypothamus-pituitary adrenal axis (de Kloet et al., 2007). However, PTSD patients exhibited a more pronounced decrease in GH secretion than TCs, as compared with HCs. Furthermore, sleep fragmentation was only observed in PTSD patients. This may suggest that GH secretion is related to stress and trauma exposure and thus altered in all trauma exposed individuals. In patients with PTSD, nocturnal GH secretion may be further compromised by sleep fragmentation, and may add up to the alterations associated with trauma exposure. Prospective, longitudinal studies in trauma exposed individuals may further elucidate the complex relationship between neurobiological changes due to trauma exposure and PTSD related alterations.

Sleep fragmentation, in the absence of a shorter TST or SWS duration, was found to predict memory performance independently of GH secretion. This is in line with a previous study that found decreased hippocampal activation and decreased performance after a night of experimentally induced sleep fragmentation (van der Werf et al., 2009). To the best of our knowledge, this is the first study that found a relationship between spontaneous sleep fragmentation in the first half of the night and decreased memory performance.

GH is secreted by the anterior pituitary into the blood. In the central nervous system GH receptors are present in the hippocampus, amygdala, and hypothalamus (Lai et al., 1991). In addition, peripheral GH influences brain function by stimulating insulin-like growth factor –I (IGF-I) release in the liver, which crosses the blood brain barrier and affects neuroplasticity by preventing apoptosis (Frago et al., 2002). In men, 60 to 70% of the daily GH secretion occurs during the first half of sleep. During sleep deprivation and interrupted sleep the GH peak is blunted or absent (Van Cauter et al., 1992). However, increased daytime and pre sleep GH secretion has been observed when nocturnal GH peaks are inhibited due to sleep restriction or circadian rhythm shifts, (Mendelson et al., 1983; Brandenberger and Weibel, 2004). The absence of a GH peak during sleep may be

a mediator in the association between sleep quality and memory consolidation. The effect of GH on hippocampal neurons during sleep deprivation has been shown in a recent preclinical study (Kim et al., 2010). This study shows that during sleep deprivation, when GH secretion was absent, N-methyl-D-aspartate (NMDA) receptor mediated synaptic currents decrease in hippocampal neurons. Moreover, NMDA receptor loss was observed during sleep deprivation, as was a decline in long term potentiation. These processes normalized when GH injections were administered during sleep deprivation. In our study we found that participants with more awakenings and less GH secretion during the night recalled fewer words on the 15 word task during a delayed recall in the morning after sleep. This may indicate that sleep dependent memory consolidation is disturbed in PTSD due to decreased nocturnal GH secretion and interrupted sleep in the first half of the night. Evidence from studies on cognitive deterioration in patients with GH hormone deficiency (GHD) further supports the relationship between GH levels and memory function (Deijen et al., 1996; van Dam et al., 2000). Moreover, placebo-controlled trials with recombinant human growth hormone (rHGH) therapy show improvement of cognitive functioning in GHD after GH administration (Oertel et al., 2004). However, our findings contrast with a study in healthy subjects that showed unaffected memory consolidation at 0300h (4-5 hours after encoding) after somatostatin administration, which inhibits GH secretion (Gais et al., 2006). The time between encoding and delayed recall was longer in our study, as we tested memory consolidation in the morning following sleep. This may have enhanced the sensitivity for detecting the influence of GH secretion on memory consolidation, as memories may have further consolidated after 0300h in the presence of GH.

In the present study, GH secretion was related to awakenings. In OSAS the same relation between awakenings and GH has been suggested. However short sleep interruptions in OSAS are accompanied by hypoxia, which also affects GH secretion; at least in preclinical studies (Zhang and Du, 2000). In our study, hypoxia does not seem to have affected GH secretion as PTSD patients exhibited equal apnea - hypopnea indexes as controls, and all participants with OSAS were excluded.

Sleep interruptions may have been a causative factor for decreased GH levels in PTSD patients. This is supported by a previous study that showed that GH secretion upon growth hormone releasing hormone (GHRH) administration was dependent on whether a subject was asleep or awake; awakenings during sleep inhibited GH secretion (Van Cauter et al., 1992). Alternatively, sleep fragmentation and decreased GH secretion may be epiphenomena and related through joined regulatory mechanisms. Several neuropeptides from the hypothalamus, such as GHRH, ghrelin and neuropeptide Y (NPY) regulate both GH secretion and non rapid eye movement (NREM) sleep. Alterations in secretion levels of these hormones may lead to a more interrupted NREM sleep and decreased GH secretion. Interestingly, low levels of NPY have been reported in PTSD and in TCs (Rasmusson et al., 2000; Morgan et al., 2003; Yehuda et al., 2006; Sah et al., 2009). Another important regu-

latory system for GH secretion is the sympathetic nervous system. Noradrenergic activity regulates GH secretion by stimulating α_2-receptors on GHRH neurons in the hypothalamus and induces GHRH release and subsequent GH secretion. In case of noradrenergic overdrive, as has been reported in PTSD, α_2-receptors become down regulated, leading to blunted GH peaks upon noradrenaline stimulation (Southwick et al., 1999; Morris et al., 2004). During sleep noradrenaline levels diminish (Lechin et al., 2004; Rasch et al., 2007), while GH levels rise. Therefore, the role of noradrenaline in nocturnal GH regulation is unclear. However, -theoretically- increased noradrenergic activity may lead to increased daytime GH secretion with as a result inhibited levels at night (Mendelson et al., 1983). One study in PTSD, showed elevated daytime GH plasma levels at three months post-trauma, at a single time-point measure (Song et al., 2008). These observations drive the need for studies to assess 24-hour GH levels in patients with PTSD.

Structural and functional changes in the hippocampus have been reported in patients with PTSD (Gurvits et al., 1996; Bremner, 2006), which are related to insomnia complaints (Neylan et al., 2010). Since GH receptors are present in the hippocampus and are involved in neurogenesis, decreased GH secretion may -theoretically- have a role in the hippocampal volume loss seen in PTSD. rHGH is on the market for the treatment of GHD and has been found to improve not only memory deficits, but also mood and quality of life (Bengtsson et al., 1993; Burman et al., 1995). Future work should indicate whether GH administration may be useful in the treatment of PTSD.

Due to a small sample size it was not possible to analyze GH secretion in PTSD with a major depressive disorder (MDD) and without MDD separately. The three patients with MDD in the present study exhibited lower values of AUCi; however, BMI and age of these patients were also somewhat higher; therefore it would be interesting to study the effect of co-morbid MDD on GH secretion in PTSD with a larger group. Furthermore, two PTSD patients that entered GH analyses used medication for hypertension. They had lower values of GH as compared to the other PTSD patients. It is unlikely that they contributed unequally to the difference with controls, as the mean of the medication naïve patients was still much lower than controls. We found statistically significant correlations between awakenings and GH secretion, and GH secretion and delayed recall in all participants together. To rule out a confounding group-effect, we performed a sub analyses in all three groups separately. The correlation between awakenings and GH secretion remained significant in HC, but not in PTSD (n=7) and TCs (n=12). This may have been due to a power problem. Regression analyses showed that both GH and awakenings significantly predicted delayed recall. We used only one declarative memory task to assess sleep dependent memory consolidation. In future studies it would be recommended to include a larger battery of memory tasks, in order to evaluate whether both neutral and emotional memory depend on sleep fragmentation and GH secretion during sleep. Due to our small sample size and the cross-sectional design of the study, we cannot conclude whether the

change in delayed recall in PTSD is the result of sleep-dependent memory consolidation or an overall worsening on memory tasks, which is nevertheless predicted by awakenings and GH secretion. To address this limitation a larger and preferably longitudinal study is warranted to replicate our preliminary findings.

In conclusion, this study shows that veterans with PTSD exhibit lower nocturnal GH plasma levels and have a more fragmented sleep. Decreased nocturnal GH secretion may be related to frequent awakenings from sleep, and both may exert a negative effect on sleep-dependent memory consolidation.

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REFERENCES

Bengtsson, B.A., Eden, S., Lonn, L., Kvist, H, Stokland, A., Lindstedt, G., Bosaeus, I., Tolli, J., Sjostrom, L., Isaksson, O.G., (1993). Treatment of adults with growth hormone (GH) deficiency with recombinant human GH. J.Clin.Endocrinol.Metab 76, 309-317.

Blake, D.D., Weathers, F.W., Nagy, L.M., Kaloupek, D.G., Gusman, F.D., Charney, D.S., Keane, T.M., (1995). The development of a Clinician-Administered PTSD Scale. J.Trauma Stress. 8, 75-90.

Brandenberger, G., Weibel, L., (2004). The 24-h growth hormone rhythm in men: sleep and circadian influences questioned. J.Sleep Res. 13, 251-255.

Belleville, G., Guay, S., and Marchand, A., (2009). Impact of sleep disturbances on PTSD symptoms and perceived health. J Nerv Ment Dis. 197, 126-32.

Bremner, J.D., (2006). Stress and brain atrophy. CNS.Neurol.Disord.Drug Targets. 5, 503-512.

Burman, P., Broman, J.E., Hetta, J., Wiklund, I., Erfurth, E.M., Hagg, E., Karlsson, F.A., (1995). Quality of life in adults with growth hormone (GH) deficiency: response to treatment with recombinant human GH in a placebo-controlled 21-month trial. J.Clin.Endocrinol.Metab 80, 3585-3590.

de Kloet, C.S., Vermetten, E., Heijnen, C.J., Geuze, E., Lentjes, E.G., Westenberg, H.G.,(2007). Enhanced cortisol suppression in response to dexamethasone administration in traumatized veterans with and without posttraumatic stress disorder. Psychoneuroendocrinology 32, 215-226.

Deijen, J.B., de Boer, H., Blok, G.J., van der Veen, e.a., (1996). Cognitive impairments and mood disturbances in growth hormone deficient men. Psychoneuroendocrinology 21, 313-322.

Frago, L.M., Paneda, C., Dickson, S.L., Hewson, A.K., Argente, J., Chowen, J.A., (2002). Growth hormone (GH) and GH-releasing peptide-6 increase brain insulin-like growth factor-I expression and activate intracellular signaling pathways involved in neuroprotection. Endocrinology 143, 4113-4122.

Gais, S., Albouy, G., Boly, M., ng-Vu, T.T., Darsaud, A., Desseilles, M., Rauchs, G., Schabus, M., Sterpenich, V., Vandewalle, G., Maquet, P., Peigneux, P., (2007). Sleep transforms the cerebral trace of declarative memories. Proc.Natl.Acad.Sci.U.S.A 104, 18778-18783.

Gais, S., Hullemann, P., Hallschmid, M., Born, J., (2006). Sleep-dependent surges in growth hormone do not contribute to sleep-dependent memory consolidation. Psychoneuroendocrinology 31, 786-791.

Gais, S., Lucas, B., Born, J., (2006). Sleep after learning aids memory recall. Learn.Mem. 13, 259-262.

Germain, A., Nielsen, T.A., (2003). Sleep pathophysiology in posttraumatic stress disorder and idiopathic nightmare sufferers. Biol.Psychiatry 54, 1092-1098.

Geuze, E., Vermetten, E., de Kloet, C.S., Hijman, R., Westenberg, H.G., (2009). Neuropsychological performance is related to current social and occupational functioning in veterans with posttraumatic stress disorder. Depress.Anxiety. 26, 7-15.

Gurvits, T.V., Shenton, M.E., Hokama, H., Ohta, H., Lasko, N.B., Gilbertson, M.W., Orr, S.P., Kikinis, R., Jolesz, F.A., McCarley, R.W., Pitman, R.K., (1996). Magnetic resonance imaging study of hippocampal volume in chronic, combat-related posttraumatic stress disorder. Biol.Psychiatry 40, 1091-1099.

Habukawa, M., Uchimura, N., Maeda, M., Kotorii, N., Maeda, H., (2007). Sleep findings in young adult patients with posttraumatic stress disorder. Biol.Psychiatry 62, 1179-1182.

Kim, E., Grover, L.M., Bertolotti, D., Green, T.L., (2010). Growth hormone rescues hippocampal synaptic function after sleep deprivation. Am.J.Physiol Regul.Integr.Comp Physiol 298, R1588-R1596.

Kobayashi, I., Boarts, J.M., Delahanty, D.L., (2007). Polysomnographically measured sleep abnormalities in PTSD: a meta-analytic review. Psychophysiology 44, 660-669.

Lahl, O., Wispel, C., Willigens, B., Pietrowsky, R., (2008). An ultra short episode of sleep is sufficient to promote declarative memory performance. J.Sleep Res. 17, 3-10.

Lai, Z., Roos, P., Zhai, O., Olsson, Y., Fholenhag, K., Larsson, C., Nyberg, F., (1993). Age-related reduction of human growth hormone-binding sites in the human brain. Brain Res. 621, 260-266.

Lai, Z.N., Emtner, M., Roos, P., Nyberg, F., (1991). Characterization of putative growth hormone receptors in human choroid plexus. Brain Res. 546, 222-226.

Lanfranco, F., Motta, G., Minetto, M.A., Ghigo, E., Maccario M., (2010). Growth hormone/insulin-like growth factor-I axis in obstructive sleep apnea syndrome: an update. J.Endocrinol.Invest 33, 192-196.

Lechin, F., Pardey-Maldonado, B., van der Dijd, B., Benaim, M., Baez, S., Orozco, B., Lechin, A.E., (2004). Circulating neurotransmitters during the different wake-sleep stages in normal subjects. Psychoneuroendocrinology, 29, 669-685.

Mellman, T.A., Kulick-Bell, R., Ashlock, L.E., Nolan, B., (1995). Sleep events among veterans with combat-related posttraumatic stress disorder. Am.J.Psychiatry 152, 110-115.

Mendelson, W.B., Jacobs, L.S., Gillin, J.C., (1983). Negative feedback suppression of sleep-related growth hormone secretion. J.Clin.Endocrinol.Metab 56, 486-488.

Morgan, C.A. III., Rasmusson, A.M., Winters, B., Hauger, R.L., Morgan, J., Hazlett, G., Southwick, S., (2003). Trauma exposure rather than posttraumatic stress disorder is associated with reduced baseline plasma neuropeptide-Y levels. Biol.Psychiatry 54, 1087-1091.

Morris, P., Hopwood, M., Maguire, K., Norman, T., Schweitzer I,. (2004). Blunted growth hormone response to clonidine in post-traumatic stress disorder. Psychoneuroendocrinology 29, 269-278.

Neylan, T.C., Mueller, S.G., Wang, Z., Metzler, T.J., Lenoci, M., Truran, D., Marmar, C.R., Weiner, M.W., Schuff, N., (2010). Insomnia severity is associated with a decreased volume of the CA3/dentate gyrus hippocampal subfield. Biol.Psychiatry 68, 494-496.

Oertel, H., Schneider, H.J., Stalla, G.K., Holsboer, F., Zihl, J., (2004). The effect of growth hormone substitution on cognitive performance in adult patients with hypopituitarism. Psychoneuroendocrinology 29, 839-850.

Pruessner, J.C., Kirschbaum, C., Meinlschmid, G., Hellhammer, D.H., (2003). Two formulas for computation of the area under the curve represent measures of total hormone concentration versus time-dependent change. Psychoneuroendocrinology 28, 916-931.

Rasch, B., Buchel, C., Gais, S., Born, J., (2007). Odor cues during slow-wave sleep prompt declarative memory consolidation. Science 315, 1426-1429.

Rasch, B., Dodt, C., Molle, M., Born, J., (2007). Sleep-stage-specific regulation of plasma catecholamine concentration. Psychoneuroendocrinology 32, 884-891.

Rasmusson, A.M., Hauger, R.L., Morgan, C.A., Bremner, J.D., Charney, D.S., Southwick, S.M., (2000). Low baseline and yohimbine-stimulated plasma neuropeptide Y (NPY) levels in combat-related PTSD. Biol.Psychiatry 47, 526-539.

Rechtschaffen, A. and Kales, A., (1968). A manual of Standardized Terminology, Techniques and Scoring System for Sleep Stages of Human Subjects. Government Printing Office, Washington DC.

Saan, R.J., Deelman, B.G., (1986). De 15 Woordentaak A en B [15 Word task]. Department of Neuropsychology, Academic Hospital Groningen, Groningen.

Sah. R., Ekhator, N.N., Strawn, J.R., Sallee, F.R., Baker, D.G., Horn, P.S., Geracioti, T.D., (2009). Low cerebrospinal fluid neuropeptide Y concentrations in posttraumatic stress disorder. Biol. Psychiatry 66, 705-707.

Song, Y., Zhou, D., Wang, X., (2008). Increased serum cortisol and growth hormone levels in earthquake survivors with PTSD or subclinical PTSD. Psychoneuroendocrinology 33, 1155-1159.

Southwick, S.M., Bremner, J.D., Rasmusson, A., Morgan, C.A.III., Arnsten, A., Charney, D.S., (1999). Role of norepinephrine in the pathophysiology and treatment of posttraumatic stress disorder. Biol.Psychiatry 46, 1192-1204.

Spitzer, R.L., Williams, J.B., Gibbon, M., First, M.B., (1992). The Structured Clinical Interview for DSM-III-R (SCID). I: History, rationale, and description. Arch.Gen.Psychiatry 49, 624-629.

Steiger, A., (2007). Neurochemical regulation of sleep. J.Psychiatr.Res. 41, 537-552.

Stickgold, R., (2005). Sleep-dependent memory consolidation. Nature 437, 1272-1278.

Takashima, A., Nieuwenhuis, I.L., Jensen, O., Talamini, L.M., Rijpkema, M., Fernandez, G., (2009). Shift from hippocampal to neocortical centered retrieval network with consolidation. J.Neurosci. 29, 10087-10093.

Van Cauter, C.E., Caufriez, A., Kerkhofs, M., Van Onderberger, A., Thorner, M.O., Copinschi, G., (1992). Sleep, awakenings, and insulin-like growth factor-I modulate the growth hormone (GH) secretory response to GH-releasing hormone. J.Clin.Endocrinol.Metab 74, 1451-1459.

Van Cauter, E., Latta, F., Nedeltcheva, A., Spiegel, K., Leproult, R., Vandenbril, C., Weiss, R., Mockel, J., Legros, J.J., Copinschi, G., (2004). Reciprocal interactions between the GH axis and sleep. Growth Horm.IGF.Res. 14 Suppl A, S10-S17.

van Dam, P.S., Aleman, A., de Vries, W.R., Deijen, J.B., van der Veen, E.A., de Haan, E.H., Koppeschaar, H.P., (2000). Growth hormone, insulin-like growth factor I and cognitive function in adults. Growth Horm.IGF.Res. 10 Suppl B, S69-S73.

Van Der Werf, Y.D., Altena, E., Schoonheim, M.M., Sanz-Arigita, E.J., Vis, J.C., De Rijke, W., Van Someren, E.J., (2009). Sleep benefits subsequent hippocampal functioning. Nat.Neurosci. 12, 122-123.

Yehuda, R., Brand, S., Yang, R.K., (2006). Plasma neuropeptide Y concentrations in combat exposed veterans: relationship to trauma exposure, recovery from PTSD, and coping. Biol.Psychiatry 59, 660-663.

Yoo, S.S., Hu, P.T., Gujar, N., Jolesz, F.A., Walker, M.P., (2007). A deficit in the ability to form new human memories without sleep. Nat.Neurosci. 10, 385-392.

Zhang, Y.S., Du, J.Z., (2000). The response of growth hormone and prolactin of rats to hypoxia. Neurosci.Lett. 279, 137-140.

Chapter 6

Pharmacotherapy for Disordered Sleep in Post-traumatic Stress Disorder: a Systematic Review

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ABSTRACT

Background. Sleep disorders, such as insomnia and nightmares, are common problems in post-traumatic stress disorder (PTSD), exert a strong negative influence on the quality of life and are a great challenge for clinical psychiatry. Several studies have reported on the efficacy of drugs for the treatment of PTSD-related sleep disorders. These studies have not been systematically reviewed. This is the first review on the effectiveness of sleep medication in PTSD.

Methods. We performed a Medline, EMBASE, and Cochrane Library Indexed search, using the keywords: PTSD, pharmacotherapy, sleep, nightmares, insomnia, and review. From this database English-language, human subject, data driven papers published after 1980 were selected.

Results. Forty eight articles are discussed. Open-label and case studies suggest efficacy for some antidepressants, anticonvulsants and atypical antipsychotics. Only a few placebo-controlled studies have been published. They show promising results for the atypical antipsychotic olanzapine, and the α_1-adrenoceptor antagonist prazosin.

Conclusions. In comparison to the incidence and impact of sleep complaints in PTSD, the pharmacotherapeutic armamentarium for PTSD-related sleep complaints remains poorly investigated. Some recent studies show promising results, especially for α_1-adrenoceptor and 5-HT_2 receptor antagonists. However, randomized controlled trials with larger populations need to be conducted.

INTRODUCTION

Post-traumatic stress disorder (PTSD) is a chronic and disabling disorder, characterized by specific symptoms that develop following exposure to trauma where the person's response involves intense fear, helplessness, or horror. Among the core symptoms of PTSD are re-experiencing of the event, avoidance of stimuli, and persistent symptoms of increased arousal. In clinical presentation of symptoms, sleep complaints are frequently reported (Geuze and Vermetten, 2004). Sleep disturbances affect about 70% of PTSD patients (Ohayon and Shapiro, 2000), with frequent nightmares and anxiety dreams, frequent awakenings, difficulty falling asleep, decreased total sleep time and restless sleep as most reported complaints (Dagan et al. 1991; Mellman et al. 1995a,b; David et al. 1996; Silva et al. 1997; Neylan et al. 1998; Schreuder et al. 1998).

Sleep disturbances warrant special attention in the treatment of PTSD symptoms, as sleep complaints are generally severe and may exist decades after the original trauma (Schreuder et al 2000). Results from controlled polysomnographic studies are not unambiguous on altered sleep latency, sleep efficiency, number of arousals, total sleep time, percentage REM and slow wave sleep, and sleep disturbed breathing (Ross et al. 1994; Mellman et al. 1995c; Dow et al. 1996; Woodward et al. 1996; Mellman et al. 1997; Engdahl et al. 2000; Woodward et al. 2000; Krakow et al. 2002; Neylan et al. 2003b; Breslau et al. 2004). A number of studies describe other alterations in sleep with unclear clinical importance. These alterations include less movement time, especially in patients with nightmares or co-morbid panic disorder (Woodward et al. 2002). Trauma-related nightmares were also associated with more wake after sleep time, whereas non-trauma-related nightmares were not (Woodward et al. 2000). Furthermore, elevated arousal thresholds during non-REM (NREM) and REM sleep have been described (Dagan et al. 1991; Lavie et al. 1998). In PTSD, more β-band power was reported during REM compared with NREM, and heart rate differences between REM and NREM were related to the amount of REM sleep (Woodward et al. 2000).

Objective criteria for impaired sleep in PTSD need further exploration in order to better understand the underlying mechanism, and to develop adequate therapeutic interventions for this severe clinical problem. From a theoretical perspective, several drugs may be useful in the treatment of disordered sleep in PTSD. Based on the reported neurobiological abnormalities in PTSD, one may argue that altered activity of noradrenalin, serotonin and gamma amino-butyric acid (GABA) may play a role in the observed sleep problems. Noradrenaline dysfunction has been reported in PTSD by several investigators (Southwick et al. 1999; Vermetten and Bremner, 2002; Charney, 2004). One study found increased sympathetic activation during REM sleep in the acute aftermath of trauma in subjects who went on to develop PTSD (Mellman et al. 2004). Additionally, an abnormal increase in methoxyhydroxyphenylglycol (the major noradrenaline metabolite) levels during

night has been observed in PTSD, which was negatively related to total sleep time (Mellman et al. 1995b). Preclinical studies show that noradrenergic activity is important for arousal regulation during sleep and for the regulation of REM sleep (Ouyang et al. 2004). Moreover, noradrenaline promotes wakefulness under stressful conditions (Hunsley and Palmiter, 2004). Agents inhibiting noradrenergic activity in the brain may thus alleviate sleep complaints. Furthermore, selective serotonin reuptake inhibitors (SSRIs) are effective for PTSD symptoms and 5-HT_2 receptors are supposed to play a critical role in SWS regulation (Idzikowski et al. 1991). In depression, 5-HT_2 receptor antagonists have shown to be effective for sleep complaints (Sharpley et al. 1994). Possibly, agents with 5-HT_2 receptor blocking properties may also have a positive influence on sleep in PTSD.

Activation of $GABA_A$ receptors plays a crucial role in the initiation and maintenance of NREM sleep (Lancel, 1999). Low plasma GABA levels have been found in patients who later developed PTSD (Vaiva et al. 2004). Moreover, decreased benzodiazepine receptor binding has been found in the prefrontal cortex of PTSD patients (Bremner et al. 2000) Also, decreased density of platelet benzodiazepine receptors has been observed in combat-related PTSD patients (Gavish et al. 1996).Thus, besides noradrenalin and serotonin, altered GABA-ergic activity may contribute to sleep disturbances in PTSD patients.

In the past two decades, several trials have been performed in which drugs were evaluated for their efficacy in the treatment of subjective sleep complaints in PTSD patients. Recently, some promising studies have been published that show new possibilities in the treatment of PTSD-related sleep complaints. The objective of this review is to provide an up-to-date overview of literature on the pharmacotherapy of sleep disorders in PTSD, and to provide suggestions for drug treatment and to formulate recommendations for future research.

MATERIALS AND METHODS

We performed a Medline, EMBASE, and Cochrane Library Indexed search using the keywords: PTSD, pharmacotherapy, sleep, nightmares, insomnia, and review. From this database English-language, human subject, data driven papers published after 1980 were selected. Articles were excluded if sleep was not evaluated and explicitly mentioned as a separate outcome measure. Forty-eight relevant articles were found. The first studies on the efficacy of pharmacotherapy for sleep disorders in PTSD were published between 1980 and 1990. The drugs studied were predominantly the tricyclic antidepressants (TCAs) and monoamine oxidase inhibitors (MAOIs). In the following decade (1990-2000) studies concentrated on the effect of SSRIs, anticonvulsants and benzodiazepines. More recently, the efficacy of (atypical) antipsychotics, the antidepressants nefazodone, trazodone and mirtazepine, and the adrenergic agents prazosin and clonidine, have been evaluated for

their efficacy in disordered sleep in PTSD. Studies will be discussed according to their classification as TCAs, MAOIs, anxiolytics and anticonvulsants, SSRIs, other antidepressants, antipsychotics and other agents.

RESULTS

Tricyclic antidepressants

TCAs are used as second-line treatment in major depressive disorder (MDD). In some countries, such as the Netherlands, TCAs are also advised as second-line treatment in PTSD. TCAs are known to reduce REM sleep, increase REM latency, and to worsen sleep continuity in the first month after treatment (Wilson and Argyropoulos, 2005). Few studies report on the use of TCAs in PTSD-related sleep complaints. One small trial compared imipramine and chloral hydrate in the treatment of 25 pediatric burn patients with acute stress disorder (Robert et al. 1999). Eighty percent of all patients treated with imipramine for 1 week experienced symptom relief compared to 45% of patients treated with chloral hydrate. Eight of the responders no longer had nightmares, and a small sample of nine experienced significant relief of insomnia. In an open label study with imipramine, in three out of the 12 PTSD patients sleep improved after 1-2 months (Kinzie and Leung, 1989).

MAO inhibitors

MAOIs are mostly used in the treatment of therapy resistant MDD. In PTSD, MAOIs have hardly been investigated. MAOIs suppress REM sleep in healthy volunteers and in MDD, whereas sleep continuity does not change (Wilson and Argyropoulos, 2005). Hogben and Cornfield (1981) reported on five veterans with PTSD treated with phenelzine in doses ranging from 45-75 mg. Remarkably, patients stopped having nightmares after 5 days to 1 month of medication. In one open-label study, recurrent dreams and sleep disturbance improved following treatment with phenelzine in 10 PTSD patients (Davidson et al. 1987). In another open-label study, 25 patients with combat related PTSD were treated with 30-90 mg of phenelzine (Lerer et al. 1987). Sleep disturbances and frequency of trauma-related nightmares decreased significantly. Only three patients dropped out due to side effects. Moclobemine improved insomnia and nightmares in an open label study in 10 combat related PTSD patients (Neal et al. 1997).

Anxiolytics and Anticonvulsants

For many years, benzodiazepines have been frequently used for the treatment of insomnia, including PTSD-related sleep complaints. Effects on sleep architecture include reduced SWS, reduced REM sleep, and increased sleep continuity (Parrino and Terzano, 1996). In a recent single-blind, placebo-controlled cross-over study, treatment of subjective sleep

disturbances with the benzodiazepine clonazepam was evaluated in 6 combat related PTSD patients (Cates et al. 2004). Each patient received clonazepam and placebo for 2 weeks, with no effect on sleep complaints during clonazepam treatment. Four case reports demonstrated that temazepam improved symptoms of acute stress, including disturbed sleep (Mellman et al. 1998). A subsequent placebo-controlled trial with temazepam in the acute aftermath of trauma revealed that subjective sleep improved in the temazepam group, but this effect was not maintained when treatment was discontinued after 7 days (Mellman et al. 2000). Moreover, temazapam was not superior to placebo in preventing PTSD symptoms. Three case studies in combat-related PTSD were reported in which insomnia and other PTSD symptoms improved with the anxiolytic buspirone (Wells et al. 1991). Case reports have suggested efficacy for zolpidem and tiagabine, both non-benzodiazepine GABAergic hypnotics, in the treatment of insomnia and nightmares in PTSD (Taylor, 2003; Dieperink and Drogemuller, 1999).

Anticonvulsants are a recent addition for treatment of disordered sleep in PTSD. Gabapentin is an anticonvulsant, which increases SWS (Legros and Bazil, 2003). In a retrospective open-label study with gabapentin addition to the standard treatment in 30 PTSD patients, an improvement for the duration of sleep was seen in 77% of the patients, and a decrease in the frequency of nightmares was noted (Hamner et al. 2001). Topiramate is another anticonvulsant that has been tested. Effects on sleep are unknown. One case study and two open-label studies suggest effect of topiramate on complaints of poor sleep in PTSD patients (Berlant, 2001; Berlant and van Kammen, 2002) (Table 1).

Table 1: Anxiolytics and anticonvulsants in the treatment of disordered sleep in post-traumatic stress disorder

Drug	Reference	Study design	Follow-up	Type of trauma	n =	Nightmares	Insomnia
Temazepam	Mellman et al. (1998)	Open-label	1 week	civilian	4	NR	↓
	Mellman et al. (2002)	RCT	1 week	civilian	22	NR	=
Buspirone	Wells et al. (1991)	Case report	3 months	combat	3	↓	↓
Zolpidem	Dieperink & Drogemuller (1999)	Case report	9 months	combat	3	↓	↓
Tiagabine	Taylor (2003)	Case report	8 weeks	civilian	7	↓	↓
Clonazepam	Cates et al. (2004)	RCT	1-2 weeks	combat	6	=	=
Gabapentin	Hamner et al. (2001)	Open-label	1-36 months	combat	30	↓	↓
Topiramate	Berlant (2001)	Case report	1-5 months	civilian	3	↓	NR
	Berlant & van Kammen (2002)	Open-label	4 weeks	civilian	35	↓	NR

↓ , Decreased; =, no change; n, number of subjects; NR, not reported.

Selective Serotonin Reuptake Inhibitors

SSRIs are commonly used in the treatment of PTSD symptoms. SSRI administration in MDD patients and healthy controls results in REM sleep decreases, REM latency increases, and worsening of sleep continuity (Wilson and Argyropoulos, 2005). Several placebo-controlled trials have been performed in PTSD patients. However, only limited studies have evaluated improvement of sleep complaints as a primary outcome measure.

Placebo-controlled trials have shown efficacy for paroxetine in the treatment of PTSD symptoms, including sleep disturbances. This was confirmed in a pooled analysis of three placebo-controlled studies (Stein et al. 2003). A placebo-controlled trial showed a significant decrease in 'trouble sleeping' after fluoxetine treatment, as measured by a self-administered questionnaire. However, the improvement was not significant as measured by a structured interview. Fluoxetine did not have an effect on nightmares in this study (Meltzer-Brody et al. 2000). In two open-label studies, the effectiveness of fluvoxamine in the treatment of sleep complaints in combat-related PTSD has been evaluated. The first study showed no significant decrease in insomnia and nightmares in 24 combat-related PTSD patients (De Boer et al. 1992). A large number of patients terminated this study prematurely, mainly due to gastrointestinal complaints. In another study, both sleep maintenance and sleep onset insomnia improved, although the effect was larger for sleep maintenance (Neylan et al. 2001). The largest effect was seen on the frequency of trauma-related dreams. Sertraline did not induce improvement in sleep quality compared to placebo (Davidson et al. 2001). In this relatively large trial (n=208) sleep was evaluated as a secondary outcome measure. Insomnia as side-effect occurred significantly more frequently in the sertraline group (35%) than in the placebo condition (22%).

Other antidepressants

Other antidepressants have not yet been tested as extensively as SSRIs in the treatment of PTSD.

Nefazodone, trazodone and mirtazepine have been examined for the treatment of sleep disorders in PTSD because their effect has been proven in treating disordered sleep in other psychiatric disorders such as MDD (Nierenberg et al. 1994; Rush et al. 1998; Manber et al. 2003; Winokur et al. 2003).

In MDD and in healthy controls nefazodone improves sleep continuity, without effects on REM sleep (Wilson and Argyropoulos, 2005). Nefazodone is a 5-HT_2 receptor antagonist, and has α_1-adrenoceptor blocking properties. Seven open-label studies on the effect of nefazodone in PTSD patients have been published (Hertzberg et al. 1998; Davidson et al. 1998; Mellman et al. 1999; Zisook et al. 2000; Gillin et al. 2001; Hertzberg et al. 2002; Neylan et al. 2003). Pooled analysis of these studies revealed significant improvement in sleep duration and reduction of nightmares (Hidalgo et al. 1999). A greater improvement was seen in patients with PTSD associated with civilian trauma, in

women, and in younger individuals. One group of authors evaluated long-term effects of nefazodone 4 years after a 12 week treatment period (Hertzberg et al. 2002). Compared to baseline (sleep quality before treatment 4 years earlier), total hours of sleep per night, and Pittsburg Sleep Quality Index (PSQI) scores were still significantly improved. However, compared to the results after 12 weeks of treatment, patients had less hours of sleep per night. All patients still met criteria for PTSD after 4 years. Three of the open-label studies evaluated the efficacy of nefazodone by using both subjective and objective measures. In the study of Mellman et al. (1999), trauma-related nightmares and total sleep time did not improve significantly in the 11 patients studied, whereas overall PTSD symptoms did. Polysomnographic (PSG) recordings obtained from four patients revealed a trend for a decrease in number of awakenings and arousals after treatment. In another study 12 male combat veterans were included (Gillin et al. 2001). Patients reported less nightmares and sleep problems after treatment with nefazodone. However, no changes in sleep architecture were seen. Finally, Neylan et al. (2003a) assessed the effectiveness of nefazodone on both subjective and objective sleep quality in 10 male veterans for 12 weeks (Neylan et al. 2003a). Subjective sleep quality and ambulatory PSG were assessed at base line and after 12 weeks. A significant improvement of insomnia complaints and a decrease in nightmares was seen. PSG showed a significant increase in total sleep time, sleep maintenance, and delta sleep after treatment with nefazodone.

Trazodone promotes sleep continuity and induces a decrease in REM sleep (Wilson and Argyropoulos, 2005). Trazodone blocks the 5-HT_{1A}, 5-$HT_{2,}$ and α_1-adrenoceptor. An open-label study with trazodone reported that sleep was the first symptom to improve in six patients with PTSD (Hertzberg et al. 1996). In a survey of 74 PTSD patients with sleep disturbances, trazodone was effective in decreasing nightmares in 72% of the 60 patients who tolerated the medication (Warner et al. 2001). Ninety-two percent of the patients considered trazodone effective with regard to sleep-onset, and 78% reported improvement in sleep maintenance. Priapism was reported as a side effect in 12% of the cases.

Mirtazapine is a α_2- adrenoceptor antagonist with H_1 and 5-HT_2 receptor antagonistic properties. It improves sleep continuity in MDD and in healthy controls (Wilson and Argyropoulos, 2005). Lewis et al (2002) published preliminary results on the effectiveness of mirtazepine. Mirtazapine was prescribed in more than 300 refugees with trauma-related nightmares. It was estimated that 75% of the patients experienced a reduction in the frequency and intensity of nightmares (Table 2).

Antipsychotics

The utility of antipsychotics in PTSD patients has been recently reviewed by Ahearn et al. 2003. In healthy subjects, olanzapine administration increases sleep continuity and enhances SWS (Sharpley et al. 2000). Additionally, olanzapine improves sleep continuity and increases SWS in patients receiving SSRI treatment (Sharpley et al. 2005). The use

Table 2: Antidepressants in the treatment of disordered sleep in post-traumatic stress disorder.

Drug	Reference	Study design	Follow-up	Type of trauma	n =	Nightmares	Insomnia	PSG
Imipramine	Robert et al (1999)	versus chloral hydrate	7 weeks	children	25	↓	↓	
	Klinzie & Leung (1989)	case report	1 year	war	12	=	=	
Phenelzine	Lerer et al (1987)	Open-label	8-18 weeks	combat	25	↓	↓	
	Hogben & Cornfield (1981)	case report	1-18 months	combat	5	↓	NR	
	Davidson et al (1987)	Open-label	4 weeks	combat	11	↓	↓	
Moclobemine	Neal et al (1997)	Open-label	12 weeks	combat	10	↓	↓	
Fluoxetine	Metzer-Brody et al (2000)	RCT	5 weeks	civilian	53	=	=	
Fluvoxamine	Neylan et al (2001)	Open-label	10 weeks	combat	21	↓	↓SM = SO	
	De Boer et al (1992)	Open-label	12 weeks	combat	24	↓	↓	
Sertraline	Davidson et al (2001)	RCT	12 weeks	civilian	208	NR	=	
Paroxetine	Stein et al (2003)	RCTs (pooled)	12 weeks	mixed	1180	NR	↓	
Nefazodone	Hidalgo et al (1999)	Open-label	12 weeks	mixed	41	↓	↓	
	Davidson et al (1998)	Open-label	12 weeks	civilian	17	↓	↓	
	Hertzberg et al (1998)	Open-label	16 weeks	combat	10	nr	↓	
	Mellman et al (1999)	Open-label	6 weeks	combat	15	↓	=	Awakenings ↓ (n = 4)
	Zisook et al (2000)	Open-label	12 weeks	combat	19	↓	↓	
	Gillin et al (2001)	Open-label	12 weeks	combat	12	↓	↓	=
	Hertzberg et al (2002)	Open-label	3-4 years	combat	10	nr	↓	
	Neylan et al (2003)	Open-label	12 weeks	combat	10	↓	↓	TST SM SWS
Trazodone	Hertzberg et al (1996)	Open-label	16 weeks	combat	74	NR	↓	
	Warner et al (2001)	Open-label	8 weeks	combat	60	↓	↓	
Mirtazapine	Lewis (2002)	Open-label	NR	war	300	↓	↓	

↓, Decreased; =, no change; NR, not reported; PSG, polysomnography; RCT, randomized controlled trial; SM, sleep maintenance; SO, sleep onset; TST, total sleep time

of olanzapine in the treatment of sleep complaints in PTSD patients has been evaluated in one serie of case reports and in one placebo-controlled study. The case reports suggested that addition of olanzapine to SSRIs is useful in treating nightmares and insomnia in patients with combat related PTSD (Jakovljevic et al. 2003). A randomized placebo-controlled trial confirmed these findings (Stein et al. 2002). Nineteen patients with PTSD were included in this double-blind, placebo-controlled study in which olanzapine was added to SSRI treatment. Although this study showed that olanzapine addition was associated with a statistically significant reduction of nightmares and improvement of insomnia, no significant global clinical improvement in PTSD symptoms was seen compared to placebo. Olanzapine use was associated with 13 lb weight gain over 8 weeks versus 3 lb weight loss in the placebo group. In a recent open-label study quetiapine alleviated nightmares and insomnia complaints in 20 combat-related PTSD patients (Robert et al. 2005). Levomepromazine was studied in a 4-week open-label study in 21 patients, and robust improvement was seen on nightmares and complaints of insomnia (Aukst-Margetic et al. 2004). Anecdotal reports show a possible efficacy for the atypical antipsychotic risperidone and thioridazine (Leyba and Wampler, 1998; Dillard et al. 1993) (Table 3).

Table 3: Antipsychotics in the treatment of disordered sleep in post-traumatic stress disorder

Drug	Reference	Study design	Follow-up	Type of trauma	n =	Night mares	Insomnia
Olanzapine	Jakovljevic et al (2003)	case report	NR	combat	5	↓	↓
	Stein et al (2002)	RCT	8 weeks	combat	19	↓	↓
Quetiapine	Robert et al (2005)	open label	6 weeks	combat	20	↓	↓
Levo-mepromazine	Aukst- Margetic et al (2004)	open label	4 weeks	combat	21	↓	↓
Risperidone	Leyba & Wampler (1998)	case report	NR	combat	4	↓	NR
Thioridazine	Dillard et al (1993)	case report	3 weeks	combat	1	↓	↓

↓, decreased; n, number of subjects; NR, not reported, RCT, randomized controlled trial

Other drugs

Prazosin, a selective α_1-adrenoceptor antagonist, is known for its antihypertensive properties, and is not widely used in the treatment of PTSD. Sleep has not been evaluated in humans after prazosin administration. In preclinical studies, duration of REM sleep per episode was blocked by prazosin in the rat (Mallick et al. 2005; Kleinlogel, 1989). In other studies prazosin facilitated REM sleep in cat (Hilakivi and Leppavuori, 1984) and in rat (Makela and Hilakivi, 1986). While treating combat-related PTSD patients with prazosin for their complaints related to benign prostate hypertrophy, the patients reported an unexpected reduction in combat-related nightmares (Raskind et al. 2000). This obser-

vation prompted the investigators to conduct an open-label feasibility trial with prazosin in five civilian PTSD patients (Taylor and Raskind, 2002) and another study in nine older men with chronic PTSD (Peskind et al. 2003). In these studies, nightmares and insomnia complaints improved. Optimal doses of prazosin ranged from 1 to 4 mg/day. Furthermore, a retrospective chart study was performed, which analysed data from 59 combat veterans with severe combat related nightmares, to whom prazosin had been prescribed (Raskind et al. 2002). The mean scores for recurrent distressing dreams improved significantly in 36 of the patients who completed at least 8 weeks of treatment. The mean maximum dose in these 36 patients was 9.6 mg per day. There were no serious adverse events attributable to prazosin. Subsequently, a small placebo-controlled cross-over study with prazosin was conducted (Raskind et al. 2003). Ten Vietnam veterans with chronic PTSD received prazosin and placebo for 9 weeks, with a wash-out period of 2 weeks in between. Prazosin was superior to placebo for treating difficulties in falling and staying asleep, for global PTSD severity, and for recurrent distressing dreams. The mean dosage of prazosin was 9.5mg/day. Seven patients used other psychoactive drugs at the time of the study. Orthostatic hypotension was reported as a transient side-effect.

Clonidine, α_2-adrenoceptor agonist, increased the amount of REM sleep and decreased the amount NREM sleep at low-dosage in healthy subjects (Miyazaki et al. 2004). By contrast, medium-dose clonidine significantly decreased REM and increased NREM. A relatively high dose of clonidine (0.3 mg/day) was effective for subjective sleep complaints in four patients (Kinzie et al. 1994). PSG showed REM suppression after administration of clonidine, however, sleep continuity did not improve. In another study, clonidine was added to imipramine in the nine patients (Kinzie and Leung, 1989). After 12-19 months of treatment, insomnia improved in six, and nightmares improved in seven out of nine patients.

Cyproheptadine, a 5-HT_2 receptor antagonist, is commonly prescribed in patients with allergic conditions. Furthermore, it is known for its sleep-promoting properties. Cyproheptadine administration is followed by a reduction in REM sleep in healthy volunteers and MDD (Sharpley et al. 1990). Several case studies have reported that cyprophepta dine is efficacious in treating nightmares in PTSD (Harsch, 1986; Brophy, 1991; Rijnders et al. 2000). In a retrospective study, nine patients with PTSD who were treated with cyproheptadine also reported remission of nightmares (Gupta et al. 1998). However, a double-blind, randomized, placebo-controlled trial in 69 patients with combat-related PTSD could not confirm these findings (Jacobs-Rebhun et al. 2000). After two weeks of treatment no improvement in sleep-related symptoms was seen in comparison to placebo.

Table 4: Prazosin, clonidine and cyproheptadine in the treatment of disordered sleep in post-traumatic stress disorder

Drug	Reference	Study design	Follow-up	Type of trauma	n =	Night mares	Insomnia
Prazosin	Raskind et al (2000)	case report	8 weeks	combat	4	↓	↓
	Taylor & Raskind (2002)	open label	6 weeks	civilian	5	↓	↓
	Raskind et al (2002)	retrospective	8 weeks	combat	59	↓	NR
	Peskind et al (2003)	open label	8 weeks	combat / holocaust	9	↓	↓
	Raskind et al (2003)	RCT	9 weeks	combat	10	↓	↓
Clonidine	Kinzie & Leung (1989)	open label	12-19 mo.	war	9	↓	↓
	Kinzie et al (1994)	case report	2 weeks	war	4	↓	↓
Cypro heptadine	Brophy et al (1991)	case report	nr	combat	4	↓	↓
	Rijnders et al (2000)	case report	1 week	civilian	1	↓	↓
	Gupta et al (1998)	case report	1 mos. - 1 yr	mixed	9	↓	NR
	Jacobs-Rebhun et al (2000)	RCT	2 weeks	combat	69	=	=

↓, decreased; = , no change; n, number of subjects; NR, not reported, RCT, randomized controlled trial

DISCUSSION

To date an insufficient number of controlled studies have been published to formulate evidence-based guidelines. Additionally, several studies have methodological limitations, such as small group sizes and heterogenic samples. Drawing on the available data, it can be concluded that TCAs are not advised as first-line treatment of PTSD-related sleep disturbances because evidence is scarce and TCAs are known to worsen sleep continuity. MAOIs are also not advised as first-line treatment of PTSD-related sleep complaints, given their potential for serious side-effects and the limited evidence for efficacy in PTSD. However, as MAOIs can completely suppress REM sleep, they may be considered in individual cases of therapy-resistant nightmares. Even though benzodiazepines are the most widely prescribed sleep medication, their effects on sleep disturbances in PTSD are generally disappointing. Clinicians should be careful in prescribing benzodiazepines because dependence and tolerance easily occur, even after short-term use. Sertraline treatment did not induce improvement in sleep; moreover, sertraline worsened sleep complaints in PTSD (Stein et al. 2002). Studies investigating the effect of paroxetine showed a different effect on sleep disturbances in PTSD compared to studies with sertraline (Davidson et al. 2001; Stein et al. 2003). Paroxetine has additional anticholinergic properties, and may be capable of suppressing REM sleep more strongly (Rao et al. 2004). Alternatively, sleep complaints may have decreased after paroxetine treatment due to the overall improvement of PTSD symptoms. In general, after SSRI treatment, residual sleep complaints are likely to occur. Other therapeutic options have to be considered in the case of persistent sleep

complaints. For example, nefazodone, trazodone and mirtazapine may have therapeutic potential in the treatment of sleep disorders in PTSD patients by virtue of their noradrenergic and $5HT_2$ receptor blocking properties. Furthermore, there is limited but promising evidence from a small placebo controlled study for treating sleep disorders in PTSD with olanzapine as add-on therapy. Possibly, other typical and atypical antipsychotics, particularly those that block the $5\text{-}HT_2$ receptors, are also effective. Weight gain is a worrisome side-effect of olanzapine. Furthermore, as in the treatment of schizophrenia, development of the metabolic syndrome in chronic atypical antipsychotic treatment in PTSD is a major concern.

The usefulness of prazosin for treating disordered sleep in PTSD seems also promising. It is relatively well evaluated in a controlled trial, and few side-effects have been reported. Based on known pharmacological properties, and supported by some open label studies, clonidine and cyproheptadine may be useful as well. Cyproheptadine was not effective in a relatively large controlled trial in chronic PTSD, but 2 weeks of treatment may be rather short to induce improvement in chronically disordered sleep.

These results should be reconfirmed in placebo-controlled trials. Furthermore, drugs should be evaluated with PSG to understand the effect on sleep architecture in PTSD. Several confounding factors must be taken into account regarding these preliminary results. The response to placebo was large in a larger randomized-controlled trial (Davidson et al. 2001). Thus, improvement in open-label studies may be due to the placebo effect. Furthermore, samples differed between studies with respect to type of trauma, age, and concurrent treatment, which limits the ability to generalize results. Also, co-morbidity, such as major depression, panic disorder and alcohol abuse, was commonly seen in most, but not all studies. Panic disorder has been shown to influence subjective sleep in PTSD (Leskin et al. 2002), and to influence frequency of nightmares and movement during sleep (Woodward et al. 2002). MDD as comorbid disorder has been shown to alter sleep functions in PTSD (Woodward et al. 1996; Mellman et al. 1997). Possibly, the occurrence of co-morbid disorders in PTSD influences response to medication. Finally, cyproheptadine, mirtazapine, nefazodone and olanzapine have sedating effects through their antihistaminergic properties, which may influence the subjective impression of sleep quality without necessarily treating underlying sleep disturbances. This finding is especially interesting in the context of the observed discrepancy between subjective and objective sleep quality in PTSD (Breslau et al., 2004; Dagan et al., 1997; Hurwitz et al., 1998; Woodward et al., 1996). Subjective complaints may improve without objective sleep quality being disturbed in the first place. In two of the three studies that correlated objective and subjective disturbances before and after nefazodone treatment, improvement of objective and subjective sleep was not in agreement. Subjective improvement and an increase in delta sleep, total sleep time, and a decrease in number of awakenings was seen in 10 patients in only one study. This study may be more reliable because ambulatory PSG at the patients'

homes was used. In all studies showing a discrepancy between subjective and objective sleep parameters in PTSD, sleep was recorded in a sleep laboratory, possibly giving a false view on the disturbed sleep in the homes of patients.

Another explanation for the discrepancy of subjective sleep and objective sleep disturbances in PTSD is that sleep disturbances may be subtle and not detectable by standard visual PSG analyses. This is suggested by two studies that found disturbances in quantitative delta activity, with normal amount of slow wave sleep (Neylan et al. 2003a; Woodward et al. 2002). Similarly, in 'sleep state misperception insomnia' or 'subjective insomnia', more subtle differences in EEG spectral activity was seen compared with subjects with normal sleep, while sleep architecture was normal according to PSG recordings (Krystal et al. 2002). Another study reported increased oxygen use during sleep in patients with subjective insomnia (Bonnet and Arand, 1997). Thus, even in the absence of disturbed sleep according to PSG, sleep may be disturbed in more subtle and yet to be explored ways.

Interestingly, PTSD-related nightmares were associated with physical sleep disorders in several studies. Patients with a history of sleep apneas reported nightmares more frequently (Groen et al 1993). Furthermore, a reduction in nightmares was observed following treatment for sleep apneas with continuous positive airway pressure. Several hypotheses may explain this finding. Hypercapnia can induce anxiety in predisposed subjects (Griez et al 1987). Furthermore, disorientation and confusion during arousals may contribute to the occurrence of nightmares after obstructive sleep apneas in PTSD patients. It has also been suggested that undiagnosed physical sleep disorders may exacerbate PTSD symptoms through chronic sleep fragmentation. Polysomnographic studies are warranted to further characterize the relation between sleep apneas, the occurrence of nightmares and PTSD severity.

Prazosin alleviates nightmare and insomnia complaints, which contributes to the hypotheses of increased noradrenergic activity in PTSD-related sleep disturbances. Olanzapine, quetiapine, nefazodone and trazodone share the α_1-adrenoceptor blocking properties with prazosin, which may explain their efficacy in this respect. Furthermore, nefazodone, mirtazepin, trazodone, risperidone and olanzapine, all have 5-HT_2 receptor blocking properties, which also may explain their efficacy in sleep disorders because 5-HT_2 receptors are implicated in sleep regulation. However, cyproheptadine, a non-selective 5-HT_2 receptor antagonist, did not improve sleep after 2 weeks of treatment in a placebo controlled trial in 69 chronic combat related PTSD patients. Most agents with positive effects are known to reduce REM sleep in MDD and healthy controls (nefazodone, trazodone, cyproheptadine, prazosine, clonidine, MAOIs, TCAs). Whether response is related to a decrease in REM sleep needs to be examined further.

In conclusion, α_1-adrenoceptor antagonists and 5-HT_2 receptor antagonists appear to be promising in the treatment of PTSD-related sleep complaints. To further develop

adequate therapeutic interventions, large randomized placebo-controlled studies need to be performed. Objective parameters for insomnia and trauma-related nightmares need to be identified for understanding underlying mechanisms of disturbed sleep in PTSD, and for evaluating therapy.

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REFERENCE LIST

Ahearn EP, Krohn A, Connor KM, and Davidson JR (2003). Pharmacologic treatment of posttraumatic stress disorder: a focus on antipsychotic use. Ann Clin Psychiatry 15:193-201.

Aukst-Margetic B, Margetic B, Tosic G, and Bilic-Prcic A (2004). Levomepromazine helps to reduce sleep problems in patients with PTSD. Eur Psychiatry 19:235-6.

Berlant JL (2001). Topiramate in posttraumatic stress disorder: preliminary clinical observations. J Clin Psychiatry 62 Suppl 17:60-3.

Berlant J, van Kammen DP (2002). Open-label topiramate as primary or adjunctive therapy in chronic civilian posttraumatic stress disorder: a preliminary report. J Clin Psychiatry 63:15-20.

Bonnet MH, Arand DL (1997). Physiological activation in patients with Sleep State Misperception. Psychosom Med 59:533-40.

Breslau N, Roth T, Burduvali E, Kapke A, Schultz L and Roehrs T (2004). Sleep in lifetime post-traumatic stress disorder: a community-based polysomnographic study. Arch Gen Psychiatry 61:508-16.

Brophy MH (1991). Cyproheptadine for combat nightmares in post-traumatic stress disorder and dream anxiety disorder. Mil Med 156:100-1.

Cates ME, Bishop MH, Davis LL, Lowe JS and Woolley TW (2004). Clonazepam for Treatment of Sleep Disturbances Associated with Combat-Related Posttraumatic Stress Disorder. Ann Pharmacother 38:1395-9.

Charney DS (2004). Psychobiological mechanisms of resilience and vulnerability: implications for successful adaptation to extreme stress. Am J Psychiatry 161:195-216.

Dagan Y, Lavie P, and Bleich A (1991). Elevated awakening thresholds in sleep stage 3-4 in war-related post-traumatic stress disorder. Biol Psychiatry 30:618-22.

David D, Mellman TA, Mendoza LM, Kulick-Bell R, Ironson G. and Schneiderman N (1996). Psychiatric morbidity following Hurricane Andrew. J Trauma Stress 9:607-12.

Davidson J, Walker JI and Kilts C (1987). A pilot of phenelzine in the treatment of post-traumatic stress disorder. Br J Psyiatry 150:252-5

Davidson JR, Rothbaum BO, van der Kolk BA, Sikes CR and Farfel GM (2001). Multicenter, double-blind comparison of sertraline and placebo in the treatment of posttraumatic stress disorder. Arch Gen Psychiatry 58:485-92.

De Boer M, Op den Velde W, Falger PJ, Hovens JE, De Groen JH, and Van Duijn H (1992). Fluvoxamine treatment for chronic PTSD: a pilot study. Psychother Psychosom 57:158-63.

Dieperink ME and Drogemuller L (1999). Zolpidem for insomnia related to PTSD. Psychiatr Serv 50:421

Dillard ML, Bendfeldt F and Jernigan P (1993). Use of thioridazine in post-traumatic stress disorder. South Med J 86:1276-8.

Dow BM, Kelsoe JR Jr and Gillin JC (1996). Sleep and dreams in Vietnam PTSD and depression. Biol Psychiatry 39:42-50.

Engdahl BE, Eberly RE, Hurwitz TD, Mahowald MW and Blake J (2000). Sleep in a community sample of elderly war veterans with and without posttraumatic stress disorder. Biol Psychiatry 47:520-5.

Gavish M, Laor N, Bidder M, Fisher D, Fonia O, Muller U et al (1996). Altered platelet peripheral-type benzodiazepine receptor in posttraumatic stress disorder. Neuropsychopharmacology 14:181-6

Geuze, E, and Vermetten E, (2004). Disordered Sleep in Posttraumatic Stress Disorder: Clinical Presentation, Research Findings, and Implications for Treatment. In: Sleep Psychiatry. (Golbin A, Kravitz H and Keith LG, eds), New York: Taylor & Francis, 247-269

Gillin JC, Smith-Vaniz A, Schnierow B, Rapaport MH, Kelsoe J, Raimo E et al (2001). An open-label, 12-week clinical and sleep EEG study of nefazodone in chronic combat-related posttraumatic stress disorder. J Clin Psychiatry 62:789-96.

Griez EJ, Lousberg H, van den Hout MA and van der Molen GM (1987). CO2 vulnerability in panic disoder. Psychiatry Res 20:87-95

de Groen JH, Op den Velde W, Hovens JE, Falger PR, Schouten EG and van Duijn H (1993). Snoring and anxiety dreams. Sleep 16:35-6.

Gupta S, Popli A, Bathurst E, Hennig L, Droney T and Keller P (1998). Efficacy of cyproheptadine for nightmares associated with posttraumatic stress disorder. Compr Psychiatry 39:160-4.

Hamner MB, Brodrick PS and Labbate LA (2001). Gabapentin in PTSD: a retrospective, clinical series of adjunctive therapy. Ann Clin Psychiatry 13:141-6.

Harsch HH (1986). Cyproheptadine for recurrent nightmares. Am J Psychiatry 143:1491-2.

Hertzberg MA, Feldman ME, Beckham JC and Davidson JR (1996). Trial of trazodone for posttraumatic stress disorder using a multiple baseline group design. J Clin Psychopharmacol 16:294-8.

Hertzberg MA, Feldman ME, Beckham JC, Moore SD and Davidson JR (1998). Open trial of nefazodone for combat-related posttraumatic stress disorder. J Clin Psychiatry 59:460-4.

Hertzberg MA, Feldman ME, Beckham JC, Moore SD and Davidson JR (2002). Three- to four-year follow-up to an open trial of nefazodone for combat-related posttraumatic stress disorder. Ann Clin Psychiatry 14:215-21.

Hidalgo R, Hertzberg MA, Mellman T, Petty F, Tucker P, Weisler R,et al (1999). Nefazodone in posttraumatic stress disorder: results from six open-label trials. Int Clin Psychopharmacol 14:61-8.

Hilakivi I and Leppavuori A (1984). Effects of methoxamine, and alpha-1 adrenoceptor agonist, and prazosin, an alpha-1 antagonist, on the stages of the sleep-waking cycle in the cat. Acta Physiol Scand 120:363-72.

Hogben GL and Cornfield RB (1981). Treatment of traumatic war neurosis with phenelzine. Arch Gen Psychiatry 38:440-5.

Hunsley MS and Palmiter RD (2004). Altered sleep latency and arousal regulation in mice lacking norepinephrine. Pharmacol Biochem Behav 78:765-73.

Idzikowski C, Mills FJ and James RJ (1991). A dose-response study examining the effects of ritanserin on human slow wave sleep. Br J Clin Pharmacol 31:193-6.

Jacobs-Rebhun S, Schnurr PP, Friedman MJ, Peck R, Brophy M and Fuller D (2000). Posttraumatic stress disorder and sleep difficulty. Am J Psychiatry 157:1525-6.

Jakovljevic M, Sagud M and Mihaljevic-Peles A (2003). Olanzapine in the treatment-resistant, combat-related PTSD--a series of case reports. Acta Psychiatr Scand 107:394-6; discussion 396.

Kinzie JD and Leung P (1989). Clonidine in Cambodian patients with posttraumatic stress disorder. J Nerv Ment Dis 177:546-50.

Kinzie JD, Sack RL and Riley CM (1994). The polysomnographic effects of clonidine on sleep disorders in posttraumatic stress disorder: a pilot study with Cambodian patients. J Nerv Ment Dis 182:585-7.

Kleinlogel H (1989). Effects of the selective alpha 1-adrenoceptor blocker prazosin on EEG sleep and waking stages in the rat. Neuropsychobiology 21:100-3.

Krakow B, Melendrez D, Johnston L, Warner TD, Clark JO, Pacheco M, et al (2002). Sleep-disordered breathing, psychiatric distress, and quality of life impairment in sexual assault survivors. J Nerv Ment Dis 190:442-52.

Krakow B, Lowry C, Germain A, Gaddy L, Hollifield M, KossM, et al (2000). A retrospective study on improvements in nightmares and post-traumatic tress disorder following treatment for co-morbid sleep-disordered breathing. J Psychosom Res 49:291-8.

Krystal AD, Edinger JD, Wohlgemuth WK and Marsh GR (2002). NREM sleep EEG frequency spectral correlates of sleep complaints in primary insomnia subtypes. Sleep 25:630-40.

Lancel M (1999). Role of GABAA receptors in the regulation of sleep: initial sleep responses to peripherally administered modulators and agonists. Sleep 22:33-42

Lavie P, Katz N, Pillar G and Zinger Y (1998). Elevated awaking thresholds during sleep: characteristics of chronic war-related posttraumatic stress disorder patients. Biol Psychiatry 44:1060-5.

Legros B and Bazil CW (2003). Effects of antiepileptic drugs on sleep architecture: a pilot study. Sleep Med 4:51-5.

Lerer B, Bleich A, Kotler M, Garb R, Hertzberg M and Levin B (1987). Posttraumatic stress disorder in Israeli combat veterans. Effect of phenelzine treatment. Arch Gen Psychiatry 44:976-81.

Leskin GA, Woodward SH, Young HE and Sheikh JI (2002). Effects of comorbid diagnoses on sleep disturbance in PTSD. J Psychiatr Res 36, 449-52.

Lewis JD (2002). Mirtazapine for PTSD nightmares. Am J Psychiatry 159:1948-9.

Leyba CM and Wampler TP (1998). Risperidone in PTSD. Psychiatr Serv 49:245-6.

Makela JP and Hilakivi IT (1986). Effect of alpha-adrenoceptor blockade on sleep and wakefulness in the rat. Pharmacol Biochem Behav 24:613-6.

Mallick BN, Singh S and Pal D (2005). Role of alpha and beta adrenoceptors in locus coeruleus stimulation-induced reduction in rapid eye movement sleep in freely moving rats. Behav Brain Res 158:9-21.

Manber R, Rush AJ, Thase ME, Amow B, Klein D, Trivedi MH et al (2003). The effects of psychotherapy, nefazodone, and their combination on subjective assessment of disturbed sleep in chronic depression. Sleep 26:130-6.

Mellman TA, David D, Kulick-Bell R, Hebding J and Nolan B (1995). Sleep disturbance and its relationship to psychiatric morbidity after Hurricane Andrew. Am J Psychiatry 152:1659-63.

Mellman TA, Kulick-Bell R, Ashlock LE and Nolan B (1995a). Sleep events among veterans with combat-related posttraumatic stress disorder. Am J Psychiatry 152:110-5.

Mellman TA, Kumar A, Kulick-Bell R, Kumar M and Nolan B (1995b). Nocturnal/daytime urine noradrenergic measures and sleep in combat-related PTSD. Biol Psychiatry 38:174-9.

Mellman TA, Nolan B, Hebding J, Kulick-Bell R and Dominguez R (1997). A polysomnographic comparison of veterans with combat-related PTSD, depressed men, and non-ill controls. Sleep 20: 46-51.

Mellman TA, Byers PM and Augenstein JS (1998). Pilot evaluation of hypnotic medication during acute traumatic stress response. J Trauma Stress 11:563-9.

Mellman TA, David D and Barza L (1999). Nefazodone treatment and dream reports in chronic PTSD. Depress Anxiety 9:146-8.

Mellman TA, Bustamante V, David D and Fins AI (2002). Hypnotic medication in the aftermath of trauma. J Clin Psychiatry 63:1183-4.

Mellman TA, Knorr BR, Pigeon WR, Leiter JC and Akay M (2004). Heart rate variability during sleep and the early development of posttraumatic stress disorder. Biol Psychiatry 55:953-6.

Meltzer-Brody S, Connor KM, Churchill E and Davidson JR (2000). Symptom-specific effects of fluoxetine in post-traumatic stress disorder. Int Clin Psychopharmacol 15:227-31

Miyazaki S, Uchida S, Mukai J and Nishihara K (2004). Clonidine effects on all-night human sleep: opposite action of low- and medium-dose clonidine on human NREM-REM sleep proportion. Psychiatry Clin Neurosci 58:138-44.

Neal LA, Shapland W and Fox C (1997). An open trial of moclobemine in the treatment of post-traumatic stress disorder. Int Clin Psychopharmacology 12:231-7.

Neylan TC, Marmar CR, Metzler TJ, Weiss DS, Zatzick DF, Delucchi KL et al (1998). Sleep disturbances in the Vietnam generation: findings from a nationally representative sample of male Vietnam veterans. Am J Psychiatry 155:929-33.

Neylan TC, Metzler TJ, Schoenfeld FB, Weiss DS, Lenoci M, Best SR, et al (2001). Fluvoxamine and sleep disturbances in posttraumatic stress disorder. J Trauma Stress 14:461-7.

Neylan TC, Lenoci M, Maglione ML, Rosenlicht NZ, Leykin Y, Metzler TJ, Schoenfeld FB and Marmar CR (2003). The effect of nefazodone on subjective and objective sleep quality in posttraumatic stress disorder. J Clin Psychiatry 64:445-50.

Neylan TC, Lenoci M, Maglione ML, Rosenlicht NZ, Metzler TJ, Otte C, et al (2003b). Delta sleep response to metyrapone in post-traumatic stress disorder. Neuropsychopharmacology 28:1666-76.

Nierenberg AA, Adler LA, Peselow E, Zornberg G and Rosenthal M (1994). Trazodone for antidepressant-associated insomnia. Am J Psychiatry 151:1069-72.

Ohayon MM and Shapiro CM (2000). Sleep disturbances and psychiatric disorders associated with posttraumatic stress disorder in the general population. Compr Psychiatry 41:469-78.

Ouyang M, Hellman K, Abel T and Thomas SA (2004). Adrenergic signaling plays a critical role in the maintenance of waking and in the regulation of REM sleep. J Neurophysiol 92:2071-82.

Parrino L and Terzano MG (1996). Polysomnographic effects of hypnotic drugs. A review. Psychopharmacology (Berl) 126:1-16.

Peskind ER, Bonner LT, Hoff DJ and Raskind MA (2003). Prazosin reduces trauma-related nightmares in older men with chronic posttraumatic stress disorder. J Geriatr Psychiatry Neurol 16:165-71.

Rao U, Lin KM, Schramm P and Poland RE (2004). REM sleep and cortisol responses to scopolamine during depression and remission in women. Int J Neuropsychopharmacol 7:265-74.

Raskind MA, Dobie DJ, Kanter ED, Petrie EC, Thompson CE and Peskind ER (2000). The alpha1-adrenergic antagonist prazosin ameliorates combat trauma nightmares in veterans with post-traumatic stress disorder: a report of 4 cases. J Clin Psychiatry 61:129-33.

Raskind MA, Thompson C, Petrie EC, Dobie DJ, Rein RJ, Hoff DJ, et al (2002). Prazosin reduces nightmares in combat veterans with posttraumatic stress disorder. J Clin Psychiatry 63:565-8.

Raskind MA, Peskind ER, Kanter ED, Petrie EC, Radant A, Thompson CE, et al (2003). Reduction of nightmares and other PTSD symptoms in combat veterans by prazosin: a placebo-controlled study. Am J Psychiatry 160:371-3.

Rijnders RJ, Laman DM and Van Duijn H (2000). Cyproheptadine for posttraumatic nightmares. Am J Psychiatry 57:1524-5.

Robert R, Blakeney PE, Villarreal C, Rosenberg L and Meyer WJ 3rd (1999). Imipramine treatment in pediatric burn patients with symptoms of acute stress disorder: a pilot study. J Am Acad Child Adolesc Psychiatry 38:873-82.

Robert S, Hamner MB, Kose S, Ulmer HG, Deitsch SE and Lorberbaum JP (2005). Quetiapine improves sleep disturbances in combat veterans with PTSD: sleep data from a prospective, open-label study. J Clin Psychopharmacol 25:387-8.

Ross RJ, Ball WA, Dinges DF, Kribbs NB, Morrison AR, Silver SM, et al (1994). Rapid eye movement sleep disturbance in posttraumatic stress disorder. Biol Psychiatry 35:195-202.

Ross RJ, Ball WA, Sanford LD, Morrison AR, Dinges DF, Silver, et al (1999). Rapid eye movement sleep changes during the adaptation night in combat veterans with posttraumatic stress disorder. Biol Psychiatry 45:938-41.

Rush AJ, Armitage R, Gillin JC, Yonkers KA, Winokur A, Moldofsky H, et al (1998). Comparative effects of nefazodone and fluoxetine on sleep in outpatients with major depressive disorder. Biol Psychiatry 44:3-14.

Schreuder BJ, van Egmond M, Kleijn WC and Visser AT (1998). Daily reports of posttraumatic nightmares and anxiety dreams in Dutch war victims. J Anxiety Disord 12:511-24.

Sharpley AL, Solomon RA, Fernando AI, da Roza Davis JM and Cowen PJ (1990). Dose-related effects of selective 5-HT2 receptor antagonists on slow wave sleep in humans. Psychopharmacology (Berl) 101:568-9.

Sharpley AL, Elliott JM, Attenburrow MJ and Cowen PJ (1994). Slow wave sleep in humans: role of 5-HT2A and 5-HT2C receptors. Neuropharmacology 33:467-71.

Sharpley AL, Vassallo CM and Cowen PJ (2000). Olanzapine increases slow-wave sleep: evidence for blockade of central 5-HT(2C) receptors in vivo. Biol Psychiatry 47:468-70.

Sharpley AL, Attenburrow ME, Hafizi S and Cowen PJ (2005). Olanzapine increases slow wave sleep and sleep continuity in SSRI-resistant depressed patients. J Clin Psychiatry 66:450-4.

Silva C, McFarlane J, Soeken K, Parker B and Reel S (1997). Symptoms of post-traumatic stress disorder in abused women in a primary care setting. J Womens Health 6:543-52.

Southwick SM, Bremner JD, Rasmusson A, Morgan CA 3rd, Arnsten A and Charney DS (1999). Role of norepinephrine in the pathophysiology and treatment of posttraumatic stress disorder. Biol Psychiatry 46:1192-204.

Stein MB, Kline NA and Matloff JL (2002). Adjunctive olanzapine for SSRI-resistant combat-related PTSD: a double-blind, placebo-controlled study. Am J Psychiatry 159:1777-9.

Stein DJ, Davidson J, Seedat S and Beebe K (2003). Paroxetine in the treatment of post-traumatic stress disorder: pooled analysis of placebo-controlled studies. Expert Opin Pharmacother 4:1829-38.

Taylor FB (2003). Tiagabine for posttraumatic stress disorder: a case series of 7 women. J Clin Psychiatry 64:1421-5.

Taylor F and Raskind MA (2002). The alpha1-adrenergic antagonist prazosin improves sleep and nightmares in civilian trauma posttraumatic stress disorder. J Clin Psychopharmacol 22:82-5.

Vaiva G, Thomas P, Ducrocq F, Fontaine M, Boss V, Devos P, et al (2004). Low posttrauma GABA plasma levels as a predictive factor in the development of acute posttraumatic stress disorder. Biol Psychiatry 55:250-4.

Vermetten E and Bremner JD (2002). Circuits and systems in stress. II. Applications to neurobiology and treatment in posttraumatic stress disorder. Depress Anxiety 16:14-38.

Warner MD, Dorn MR and Peabody CA (2001). Survey on the usefulness of trazodone in patients with PTSD with insomnia or nightmares. Pharmacopsychiatry 34:128-31.

Wells BG, Chu CC, Johnson R, Nasdahl C, Ayubi MA, Sewell E et al (1991). Buspirone in the treatment of posttraumatic stress disorder. Pharmacotherapy 11:340-3.

Wilson S and Argyropoulos S (2005). Antidepressants and sleep: a qualitative review of the literature. Drugs 65:927-47.

Winokur A, DeMartinis NA 3rd, McNally DP, Gary EM, Cormier JL and Gary KA (2003). Comparative effects of mirtazapine and fluoxetine on sleep physiology measures in patients with major depression and insomnia. J Clin Psychiatry 64:1224-9.

Woodward SH, Friedman MJ and Bliwise DL (1996). Sleep and depression in combat-related PTSD inpatients. Biol Psychiatry 39:182-92.

Woodward SH, Arsenault NJ, Murray C and Bliwise DL (2000). Laboratory sleep correlates of nightmare complaint in PTSD inpatients. Biol Psychiatry 48:1081-7.

Woodward SH, Murburg MM and Bliwise DL (2000). PTSD-related hyperarousal assessed during sleep. Physiol Behav 70:197-203.

Woodward SH, Leskin GA and Sheikh JI (2002). Movement during sleep: associations with posttraumatic stress disorder, nightmares, and comorbid panic disorder. Sleep 25:681-8.

Zisook S, Chentsova-Dutton YE, Smith-Vaniz A, Kline NA, Ellenor GL, Kodsi AB et al (2000). Nefazodone in patients with treatment-refractory posttraumatic stress disorder. J Clin Psychiatry 61: 203-8.

Chapter 7

Prazosin treatment for sleep disturbances in veterans with a post-traumatic stress disorder; a placebo-controlled pilot study using polysomnography

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ABSTRACT

Introduction. Nightmares and insomnia are highly prevalent in patients with post-traumatic Stress Disorder (PTSD), while therapeutic options are limited. Some randomized controlled trials (RCTs) suggest that prazosin, a α_1-adrenoceptor antagonist, alleviates nightmares and insomnia in PTSD. As sleep disturbances in PTSD are characterized by frequent awakenings, we hypothesized that awakenings would decrease after prazosin treatment parallel to improvement of insomnia and nightmare complaints.

Methods. We conducted a randomized double-blind placebo controlled trial in 14 PTSD patients. Subjective sleep measures, clinician administered PTSD Scale (CAPS), and polysomnographic (PSG) registration of two nights were obtained at baseline and in week 8. Sleep registrations were recorded at the participants' homes.

Results. The number of awakenings on PSG were not significantly decreased after treatment in the total study group. A reduction in the Pittsburgh Sleep Quality Index (PSQI) and CAPS scores was observed in both groups. Differences between groups were not significant. Patients receiving prazosin reported significantly more side effects.

Discussion. α_1-adrenoceptor activity was not associated with decreased awakenings. However due to small sample sizes, the power to detect treatment-related differences was limited. Larger studies are needed to support our observations.

INTRODUCTION

In the past three decades several open-label trials have studied the effect of benzodiazepines, anticonvulsants and atypical antipsychotics on sleep disturbances in patients with a post-traumatic stress disorder (PTSD) (van Liempt et al., 2006). Only a limited number of randomized controlled trials (RCT) have been published.

The discovery of prazosin in the treatment of sleep disturbances in PTSD patients was an instance of serendipity. Veterans with benign prostatic hypertrophy, who were prescribed prazosin, unexpectedly reported cessation of posttraumatic nightmares (Raskind et al., 2000). Several studies, of which were three small randomized controlled trials (RCTs), have shown positive results on subjective complaints (Germain et al., 2011; Raskind et al., 2007; Raskind et al., 2003).

Prazosin may inhibit activity of increased noradrenaline, which has been described in patients with PTSD (Baker et al., 1999; Southwick et al., 1999). Noradrenaline is a neurotransmitter known to stimulate the ascending reticular arousal system (ARAS), and inhibiting sleep-promoting ventrolateral preoptic nucleus (VLPO) activity (Saper et al., 2005). The ARAS and VLPO, and related neurotransmitters, amongst which NE, are also referred to as the "sleep/wake switch". Preclinical studies supported the hypothesis that noradrenergic activity is important for arousal regulation during sleep. Noradrenaline lacking mice exhibit elevated arousal thresholds in stressful situation (Hunsley and Palmiter, 2004). Also, decreased REM sleep duration and decreased wake time was observed in mice lacking noradrenaline (Ouyang et al., 2004).

In PTSD patients, prazosin treatment increased REM duration and total sleep time (TST) compared with a placebo in one small crossover RCT (Taylor et al., 2008). In this RCT by Taylor et al (2007) an eye tracker was used to assess eye movements to estimate REM sleep and total sleep time. However, this effect of prazosin was not seen in another RCT with a parallel design, which used polysomnography, the golden standard to determine sleep architecture (Germain et al., 2011). An increased number of awakenings may be a core feature of disturbed sleep in PTSD, while sleep architecture is relatively normal (Breslau et al., 2004; Germain et al., 2011; Habukawa et al., 2007; Kobayashi et al., 2007). Both RCTs mentioned above, assessing the effect of prazosin on objective sleep measures, did not take the number of awakenings into account. We hypothesized that prazosin would lead to reduced insomnia and nightmare symptoms, parallel to a decrease in the number of awakenings in veterans with PTSD.

MATERIALS AND METHODS

Participants

Veterans with PTSD were recruited from the four outpatient clinics of the Military Mental Health (MMH) institute in the Netherlands. Patients were invited to the Research department of the MMH in Utrecht after a telephone screening in which eligibility was tested. Of the 19 patients who were screened, 14 enrolled the study protocol. After a complete verbal and written description of the study, participants signed an informed consent. PTSD and co-morbidity were tested with a Structured Clinical Interview for DSM-IV (SCID), (Spitzer et al., 1992) and a Clinician Administered PTSD Scale (CAPS) (Blake et al., 1995). Additionally, the sleep symptoms were assessed with the Pittsburgh Sleep Quality Index (PSQI) (Buysse et al., 1989), a self-administered sleep questionnaire and an addendum of the PSQI for PTSD (Germain et al., 2005). Only patients with a score on the CAPS of 50 or higher and a PSQI of 5 or higher were included. Patients who fulfilled criteria of alcohol or substances abuse dependence were excluded. Also, patients with a history of hypertension and current use of antihypertensive medication were excluded, as prazosin lowers blood pressure. Most patients received psychotropic drugs or were in psychotherapy while entering the study. However, they did not receive medication with α_1-adrenoceptor blocking properties, and did not recently commence (within 6 weeks before the study) with psychotherapy. Participants were excluded when they needed to change psychotropic medication during the trial. All participants were between 18-65 years old. The protocol was approved by the ethical review board of the University Medical Centre Utrecht (UMCU) (2007/230). Participants were included between January 2009 and December 2011. The trial was registered at the European Clinical Trials Database;

Table 1: Inclusion and exclusion criteria.

Inclusion criteria
Age between 18 – 65
DSM-IV criteria for PTSD, as measured by SCID
CAPS score of > 50
PSQI scores of > 5
Exclusion criteria
Substance or alcohol abuse/dependence within the past six months
Major systemic or neurological diseases
Orthostatic hypotension before treatment
Use of psychotropic medication with alpha1 andrenoceptor antagonizing properties.
Use of antihypertensive agents
Psychotherapy in the last month preceeding the trial period or during the trial period, unless the patient had been receiving psychotherapy for longer than 6 weeks without improvement of sleep complaints.

EudraCT 2007-000030-39. This trial was performed according to Good Clinical Practice guidelines and was monitored externally by Julius Clinical Trial, Zeist, The Netherlands.

Study design

This was a randomized double-blind placebo-controlled trial. At study entry clinical interviews and pen and pencil questionnaires were administered, which were repeated in week 8. Furthermore, polysomnographic (PSG) registrations of two nights at the patients' homes were recorded at baseline and at week 8.

After clinical interviews and screening, patients were randomized according to a computer generalized list provided by the pharmacy of the UMCU. The pharmacy also made capsules of prazosin and placebo, which were identical in appearance.

The capsules were packaged in blisters for 4 weeks, which were dispensed twice; once for the titration period of four weeks (1 mg for four days, 2 mg for a week, 4 mg for a week, 7 mg for a week, and 10 mg for 3 days), and again for the last four weeks optimal dosage (10 or 14 mg). The dosage of 10 mg was maintained during the last four weeks when sleep complaints subsided, or when patients reported side effects. In case symptoms had not (completely) subsided, or when there were no major side-effects, the dosage was increased to 14 mg for 4 additional weeks.

A last observation-carried-forward principle was applied when participants discontinued. When participants discontinued in the titration phase (the first four weeks), no post-treatment PSG data were obtained and baseline PSG data were excluded from further analyses (prazosin n = 1, placebo n = 1).

During the treatment period patients returned to the research centre three times for a physical check-up including blood pressure measurement. At the last visit participants were asked if they thought they had been randomized to prazosin or placebo, and whether they wished to continue with the medication after the trial. The investigators also recorded whether they thought the participant had received placebo or prazosin.

Participants also filled out a side effect log during the 8 weeks of treatment. They were requested to also make a note when they missed a dosage. For an extra check of the drug accountability, participants were requested to return the 4 weeks blister in which they left the medication they missed. Before the treatment started, and in the last week of treatment, participants were requested not to drink alcohol on the days of the sleep registrations, and to not drink caffeine-containing beverages after 1600h.

Data analyses

Sleep Questionnaires

The Pittsburgh Sleep Quality Index (PSQI) and addendum of the PSQI for PTSD (Buysse et al., 1989; Germain et al., 2005) were translated into Dutch and were filled out at baseline

and in week 8 of the trial. The PSQI consists of 7 components, of which one component comprises sleep medication use. This component was left out in the total score, as all participants mentioned to have used sleep medication at week 8, referring to the study medication.

A sleep log was filled out every morning upon awakening by the participants during one week at baseline and at week 8. We calculated the mean total sleep time (MTST), mean sleep onset latency (MSOL), and mean subjective waking after sleep onset (MWASO) for the seven nights in the home situation.

After the second night of PSG at baseline and in week 8, a sleep log assessing subjective SOL, TST and WASO was also filled out on the morning. Additionally, participants marked on a visual analogue scale (VAS) how deep they perceived their sleep to be during the recording night on a 0 to 100 scale. Finally, participants were asked to grade habitual sleep in the last two weeks and sleep during the recording night with a rating between 0 and 10.

Sleep recordings

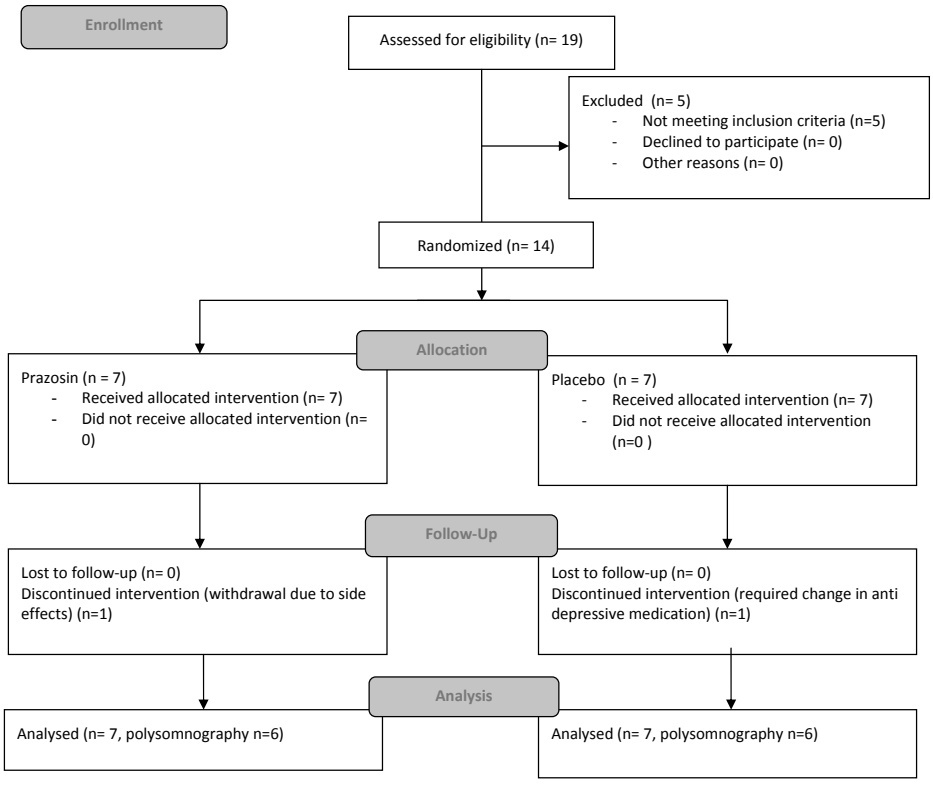
Sleep registrations during two consecutive nights were conducted at the homes of the participants. Ambulatory sleep recordings were acquired with Twente Medical Systems, Porti 7 (TMS International BV), including bipolar derivations of EMG (chin), EOG for vertical and horizontal eye movements, EEG (F0-CO, F3-C3, P3-O1, C4-A1, O2-A2) and ECG. The first night served as adaptation to the sleep registration. Participants were instructed to have a 8 hour sleep period on the registration nights. Sleep data were analyzed in 30-second epochs according to criteria of Rechtschaffen and Kales (1968) by an experienced sleep technician who was blinded to group identity. TST, WASO, and SOL, sleep stages and the number of spontaneous awakenings were determined.

Statistical analyses

Differences between the groups were tested in SPSS 17.0. Differences in demographic variables were tested with ANOVA. Due to small sample sizes most variables were not normally distributed after log-transformations, with an exception of the total CAPS score. Therefore we could not test differences between baseline and week 8 with a repeated measures ANOVA. Instead, Δ(baseline – week 8,) was calculated for all variables. An ANCOVA, correcting for the baseline value, was used to test whether deltas were statistically different between the prazosin and placebo group. For some main parameters (CAPS, B2, PSQI and number of awakenings), treatment effects in the combined sample was calculated with a repeated ANOVA (total CAPS score), or nonparametric Wilcoxon Signed Rank Test for a related sample. For dichotomous variables a chi square test was used. A nonparametric Mann-Whitney test for group differences was used for variables that were

not normally distributed after log transformation. Differences were considered significant when p values were smaller than 0.05.

Figure 1. Flow diagram



RESULTS

One participant receiving prazosin terminated the study due to side effects in the titration period. One patient receiving placebo required switching anti-depressive medication during the titration period (See figure 1).

CAPS scores and subjective sleep questionnaires of 14 participants were available for further analysis (prazosin n = 7, placebo n = 7). Polysomnographic recordings at baseline and at week 8 was available of 12 participants (prazosin n = 6, placebo n = 6). Table 2 shows demographic and medical characteristics of both groups. The groups only differed in caffeine intake, which was higher in placebo 7.42 (11.34), than in prazosin 2.43 (2.30) (Mann Whitney U = 4.50, Z=-2.58, p = 0.01).

Table 2. Demographics

Demographics	Prazosin (n=7)		Placebo (n=7)		Comparison
Age (years) (M, SD)	43.43	(5.22)	44.90	(10.53)	F (1,13) = 0.103, n.s.
Alcohol (units / week) (M, SD)	7.57	(8.34)	7.00	(8.02)	U = 22.50, Z=-0.26, n.s.
Cafeïne (cups / day) (M, SD)	2.43	(2.30)	7.42	(11.34)	U = 4.50, Z=-2.58, p = 0.01
Smoking (number of cigarettes/ day) (M, SD)	5.71	(11.34)	7.00	(8.9)	U = 21.00, Z=-0.73, n.s.
BMI (kg/l^2) (M, SD)	26.61	(2.08)	26.04	(3.60)	U = 13.00, Z=-0.26, n.s.
MDD (n, %)	1	(14%)	2	(29%)	χ^2= 0.42, df = 1, n.s.
MDD lifetime (n, %)	6	(86%)	5	(71%)	χ^2= 0.48, df = 1, n.s.
SSRI (n, %)	3	(43%)	2	(29)	χ^2= 0.31, df = 1, n.s.
Other medication (n, %) (Bupropion)	1	(14%)	0		χ^2= 1.08, df = 1, n.s.
Year First Mission (M, SD)	1990.00	(6.58)	1990.86	(8.32)	F (1,13) = 0.046, n.s.
Number of Missions (M, SD)	1.43	(0.79)	2.43	(1.40)	F (1,13) = 2.97, n.s.

BMI = Body Mass Index; M = Mean; MDD= Major Depressive Disorder; SD = Standard Deviation; SSRI = Selective Serotonin Reuptake Inhibitor

Side effects and expectancy

Significantly more participants complained of side effects in the prazosin group compared with the placebo group (see table 3). A similar number of patients in the prazosin group and placebo group reported the wish to continue medication after the trial. The expectation to have received prazosin was similar in both groups, indicating that the groups were well blinded.

Clinical interviews and sleep diary measures

CAPS score were significantly lower after treatment in the combined group (F (1,11) = 7.84, p = 0.017), however, the group x time effect was not significant (F (1,11) = 1.06, p=0.32). B2 score (nightmares) was not significantly decreased after treatment (Z = -0.97, p = 0.33). See table 4. Two patients reported normal dreaming after the start of prazosin, while they could not recall dream content at baseline. This was not reported by participants using placebo (Group difference, χ^2 = 2.33, df=1, p=0.13).

The PSQI score was lower at week 8 compared with baseline in the combined sample (Z =-2.82, p = 0.005). The decrease was not larger in patients receiving prazosin (*F* (1,10) = 0.89, *p* = 0.37). The addendum of the PSQI for PTSD was not lower after treatment in the combined sample (Z= 1.34, p=0.18). No group differences were observed in ΔMSOL, ΔMWASO and ΔMTST. See table 4 for group differences in Δ(baseline - week 8).

Polysomnography (PSG)

PSG recordings revealed no changes in the prazosin group compared with the placebo group (table 5). The number of awakenings did not significantly decrease after treatment in the total sample (Z =-1.25, p = 0.21).

Table 3. Side effects, discontinuation, expectation of treatment characteristics

	Prazosin (n=7)	Placebo (n=7)	Comparison
Missed dose (n, %)	1 (14%)	3 (43%)	$\chi^2 = 1.33$, df =1, p=0.51
Discontinuation (n,%)	1 (14%)	1 (14%)	$\chi^2 = 0.14$ df=1, p=0.91
Expectancy, participant (n, %)	Prazosin 5 (71%) Placebo 0 (0%) Unsure 2 (29%)	Prazosin 3 (43%) Placebo 3 (43%) Unsure 1 (14%)	$\chi^2 = 2.77$ df=2, p=0.25
Dosage	10 mg 3 (43%) 14 mg 3 (43%)	14 mg 7 (100%)	$\chi^2 = 4.00$ df=1, p=0.046
Expectancy, researcher (n, %)	Prazosin 3 (43%) Placebo 3 (43%) Missing = 1 (14%)	Prazosin 3 (43%) Placebo 3 (43%)	$\chi^2 = 0.11$, df =1, p=0.74
Wish to continue	No =2 (29%) Yes =2 (29%) Indecisive =2 (29%)	No =1 (14%) Yes =3 (43%) Indecisive =1 (14%)	$\chi^2 = 1.23$, df=2, p=0.54
Side effect			
Drowsiness (n, %)	5 (71%)	0 (0%)	$\chi^2 = 7.78$, df=1, p=0.005
Headache (n, %)	2 (29%)	0 (0%)	$\chi^2 = 2.33$, df=1, p=0.13
Vertigo (n, %)	5 (71%)	0 (0%)	$\chi^2 = 7.78$, df=1, p=0.005
Perspiration (n, %)	1 (14%)	1 (14%)	$\chi^2 = 0.14$ df=1, p=0.91
Other (n, %)	2 (28%)	0 (0%)	$\chi^2 = 2.33$, df=1, p=0.13

Table 4. Psychiatric symptoms and sleep questionnaires before and after treatment

Psychometric variables *(M, SD)*	Prazosin (n=7)		Placebo (n=7)		Group comparison of Δ (baseline – week 8)
	Baseline	Week 8	Baseline	Week 8	
CAPS	65.57 (15.69)	52.29(18.79)	68.43 (11.62)	62.57 (14.88)	*F(1, 12)= 1.06 p=0.32*
B2	3.71 (2.69)	2.71 (1.90)	3.50 (2.74)	3.67 (1.97)	*F(1,12) = 1.25, p=0.29*
D1	6.57 (0.79)	4.57 (1.51)	7.29 (0.76)	5.86 (1.57)	*F(1, 12) = 0.53, p=0.48*
B	18.71 (4.39)	12.14 (6.31)	17.29 (4.96)	17.86 (5.84)	*F(1, 12)= 4.27, p=0.061*
C	20.86 (9.69)	18.14 (11.34)	22.83 (5.64)	21.33 (4.23)	*F(1, 12) = 0.22, p=0.65*
D	26.00 (5.39)	22.00 (5.72)	27.50 (2.88)	21.17 (5.27)	*F(1, 12) = 0.35, p=0.57*
HAM-D	9.14 (3.93)	9.29 (6.29)	11.00 (5.62)	9.83 (2.71)	*F(1, 12) = 0.33, p=0.58*
Sleep questionnaires					
PSQI	13.50 (4.09)	8.33 (2.50)	13.17(1.47)	9.83 (3.97)	*F(1, 9) = 0.84, p=0.38*
PSQI, PTSD	8.80 (5.89)	6.00 (4.00)	8.83 (2.23)	6.83 (3.43)	*F(1, 8) = 0.12, p=0.74*
MTST (minutes)	366.15 (51.75)	405.00 (44.55)	347.55 (104.7)	352.65 (88.60)	*F(1, 8) = 0.48, p=0.51*

Table 4. (continued)

Psychometric variables *(M, SD)*	Prazosin (n=7)		Placebo (n=7)		Group comparison of Δ (baseline – week 8)
	Baseline	Week 8	Baseline	Week 8	
MWASO (minutes)	69.90 (40.65)	35.40 (29.10)	89.10 (31.65)	57.30 (59.70)	*F(1, 8) = 0.26, p=0.63*
MSOL (minutes)	51.90 (28.95)	30.30 (23.25)	53.55 (59.85)	39.90 (44.25)	*F(1, 10) = 0.34, p=0.58*

CAPS = Clinician administered PTSD Scale, HAM-D – Hamilton depression, PSQI = Pittsburgh sleep Quality Index, PSQI-PTSD = PSQI addendum for PTSD, MTST = Mean total sleep time, MWASO = mean wake after sleep onset, MSOL = mean sleep onset latency.

Table 5: Recording night; polysomnography and subjective sleep quality

Subjective Sleep Measurements *(M, SD)*	Prazosin (n=6)		Placebo (n=6)		Group comparison of Δ (baseline – week 8)
	Baseline	Week 8	Baseline	Week 8	
TST (minutes)	325.00 (61.97)	367.50 (59.06)	317.50 (99.69)	347.20 (95.54)	*F (1, 9) = 0.15, p=0.71*
WASO (minutes)	75.00 (2.71)	37.50 (33.87)	132.50(65.33)	72.50 (100.59)	*F (1, 9) = 0.21, p=0.66*
SOL (minutes)	40.00 (26.26)	25.00 (15.49)	35.00 (40.99)	25.00 (40.99)	*F (1, 9) = 0.01, p=0.98*
Sleep depth (VAS 0 -100)	54.77 (18.39)	70.9 (6.37)	36.82 (21.12)	54.62 (21.69)	*F (1, 9) = 1.59, p=0.24*
Polysomnography					
Time in bed (minutes)	431.80 (106.60)	426.60 (66.45)	454.00 (75.35)	436.73 (43.68)	*F (1, 9) = 0.13, p=0.73*
TST (minutes)	378.25 (78.48)	398.17 (50.42)	411.58 (41.39)	389.83 (59.01)	*F (1, 9) = 1.54, p=0.25*
WASO (minutes)	41.67 (59.72)	35.08 (22.79)	41.08 (48.36)	34.83 (20.38)	*F (1, 9) = 0.00, p=0.99*
S1 (% of TST)	7.28 (3.19)	7.00 (1.94)	5.42 (0.96)	6.00 (3.99)	*F (1, 9) = 0.01 p=0.97*
S2 (% of TST)	48.32 (8.78)	48.75 (7.55)	55.08 (3.41)	54.95 (7.42)	*F (1, 9) = 0.05, p=0.82*
S3 (% of TST)	9.35 (3.76)	9.02 (3.66)	8.03 (1.32)	8.27 (2.96)	*F (1, 9) = 0.28, p=0.61*
S4 (% of TST)	12.15 (3.78)	8.58 (5.96)	6.10 (3.73)	7.32 (6.70)	*F (1, 9) = 1.71, p=0.22*
REM (% of TST)	22.88 (7.12)	26.67 (3.06)	25.37 (1.59)	23.48 (5.52)	*F (1, 9) = 0.84, p=0.38*
No. Awakenings	20.33 (12.43)	16.67 (12.43)	16.50 (8.55)	12.83 (11.65)	*F (1, 9) = 0.12, p=0.73*

M= mean, No. = Number, REM = Rapid eye movement sleep, SD = Standard deviation, SOL = Sleep onset latency, TST = Total sleep time, WASO = Wake after sleep onset

DISCUSSION

In this study we used subjective sleep questionnaires and polysomnography (PSG) to assess the effect of prazosin on sleep in PTSD patients. Prazosin (n=6) was not more effective than placebo (n=6) with respect to PSG, Pittsburgh sleep quality index (PSQI) and sleep diary measurements. Prazosin led to significantly more side effects.

Re-experiencing symptoms were reduced in the prazosin groups compared with the placebo group at a trend level (p=0.061). However, due to multi-testing of CAPS scores, significant p values were below $p < 0.008$ (Bonferroni corrected). CAPS scores were significantly lower after post-treatment in prazosin group ($F(1,6)= 10.2$, $p=0.019$), but were not in the placebo group ($F(1,5) = 1.42$, $p=0.29$). However, no group differences in Δ(baseline – post-treatment) between prazosin and placebo were observed. Even though we did not observe group differences in nightmares, CAPS scores and re-experiencing symptoms, it is not to say the differences do not exist. Our small study (14) was only capable of detecting large effect sizes (> 0.82).

Still, our findings are in agreement with a recent RCT with three groups (prazosin, cognitive behavior therapy focused on sleep, and placebo), which showed that all three conditions led to improvement of PSG measures, sleep diaries and the Pittsburgh sleep quality index (PSQI) (Germain et al., 2011). There were no time x group interactions for PSG parameters, and sleep diary measures. An exception was the effect on nightmares, which only improved in the two active treatment groups. In our study too, nightmare score was somewhat reduced solely after prazosin treatment, however non-significantly. The calculated effect size of the group difference was 0.56, meaning that a total sample size of 27 was needed to find a significant group difference ($\alpha = 0.05$, $\beta= 0.80$).

We expected that with the blocking of noradrenaline activity awakenings would decrease, while noradrenaline is involved in the regulation of sleep and waking, and is well known as a wake-promoting neurotransmitter (Saper et al., 2005). In PTSD, sleep fragmentation has been previously reported, and was also seen in our study at baseline in both groups. To our surprise, awakenings at week 8 were not significantly different from baseline. When this finding would be replicated in larger samples, it would suggest that α_1 receptor activity may not be related to sleep fragmentation in PTSD.

One study contrasts with our observations. In this RCT with a crossover design (n=10) a "REMView" eye tracker was applied at baseline, after the first treatment period (with prazosin or placebo) and after the second treatment period (with prazosin or placebo) (Taylor et al., 2008). Prazosin was associated with increased TST and REM sleep. Awakenings were not assessed, and NREM sleep could not be distinguished with this technique. It is unclear why this study found differences in TST and REMS while our study and the study by Germain et al (2011) did not. An Eye Tracker is less reliable for measuring sleep architecture than PSG. Secondly, a cross sectional design may be less "double blind".

When a person has experienced side effects and/or improvement in the first treatment period, he or she may be biased in the following period.

PSG allows us to determine the architecture of sleep in sleep stages but does not provide information on brain activity in for instance limbic structures such as the amygdala during sleep (Germain et al., 2008). Nightmares may be related to increased activity of limbic structures during sleep (Nielsen and Levin, 2007). An increase in activity of limbic structures does not have to lead to changes in the sleep architecture as measured by PSG. Pharmacological agents may alleviate (or induce) nightmares, without altering sleep architecture. An example is propranolol, which can cause nightmares, but does not seem to influence sleep architecture (Smith et al., 2006). PSG may not be the method of choice when investigating mechanism through which prazosin influences sleep complaints. Brain imaging during sleep may elucidate neurobiological processes underlying nightmares in PTSD.

The results of our study should be interpreted cautiously. Firstly, the sample sizes were small. We therefore lacked the power to find group differences with effect sizes smaller than 0.82. Also, significant baseline differences were present between groups, despite randomization of the subjects. For instance, coffee intake was significantly higher in the placebo group. In larger samples is it less likely to have significantly different baseline values. Another limitation may have been the home sleep registrations. Even though it is a more naturalistic way to record sleep, there was no surveillance on time spend in bed, or coffee and medication usage. Patients received clear instructions, but we saw on the sleep registrations that 'time in bed' deviated from 8 hours in some participants.

In summary, our study failed to detect treatment-related differences after prazosin treatment, compared with placebo. This might have been due to a small sample size. On the other hand, a larger recent RCT with polysomnography also suggested that improvement of PSG parameters and subjective sleep complaints were unspecific treatment effects. In contrast, inhibition of α_1-adrenoceptor mediated activity may specifically affect re-experiencing symptoms, including nightmares.

In future research other methods than PSG, for instance combined functional MRI and EEG, may be useful to study a possible relationship between increased amygdala activity, which cannot be evaluated with PSG, and nightmares. A simultaneous fMRI/EEG study combined with prazosin administration may further elucidate the effect of blocking α_1-adrenoceptor activity on nightmares in PTSD.

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REFERENCE LIST

Baker DG, West SA, Nicholson WE, Ekhator NN, Kasckow JW, Hill KK, Bruce AB, Orth DN, Geracioti TD, Jr. (1999). Serial CSF corticotropin-releasing hormone levels and adrenocortical activity in combat veterans with posttraumatic stress disorder. Am J Psychiatry 156:585-588.

Blake DD, Weathers FW, Nagy LM, Kaloupek DG, Gusman FD, Charney DS, Keane TM (1995). The development of a Clinician-Administered PTSD Scale. J Trauma Stress 8:75-90.

Breslau N, Roth T, Burduvali E, Kapke A, Schultz L, Roehrs T (2004). Sleep in lifetime posttraumatic stress disorder: a community-based polysomnographic study. Arch Gen Psychiatry 61:508-516.

Buysse DJ, Reynolds CF, III, Monk TH, Berman SR, Kupfer DJ (1989). The Pittsburgh Sleep Quality Index: a new instrument for psychiatric practice and research. Psychiatry Res 28:193-213.

Germain A, Buysse DJ, Nofzinger E (2008). Sleep-specific mechanisms underlying posttraumatic stress disorder: integrative review and neurobiological hypotheses. Sleep Med Rev 12:185-195.

Germain A, Hall M, Krakow B, Katherine SM, Buysse DJ (2005). A brief sleep scale for Posttraumatic Stress Disorder: Pittsburgh Sleep Quality Index Addendum for PTSD. J Anxiety Disord 19:233-244.

Germain A, Richardson R, Moul DE, Mammen O, Haas G, Forman SD, Rode N, Begley A, Nofzinger EA (2011). Placebo-controlled comparison of prazosin and cognitive-behavior treatments for sleep disturbances in US Military Veterans. J Psychosom Res.

Habukawa M, Uchimura N, Maeda M, Kotorii N, Maeda H (2007). Sleep findings in young adult patients with posttraumatic stress disorder. Biol Psychiatry 62:1179-1182.

Hunsley MS, Palmiter RD (2004). Altered sleep latency and arousal regulation in mice lacking norepinephrine. Pharmacol Biochem Behav 78:765-773.

Kobayashi I, Boarts JM, Delahanty DL (2007). Polysomnographically measured sleep abnormalities in PTSD: a meta-analytic review. Psychophysiology 44:660-669.

Nielsen T, Levin R (2007). Nightmares: a new neurocognitive model. Sleep Med Rev 11:295-310.

Ouyang M, Hellman K, Abel T, Thomas SA (2004.) Adrenergic signaling plays a critical role in the maintenance of waking and in the regulation of REM sleep. J Neurophysiol 92:2071-2082.

Raskind MA, Dobie DJ, Kanter ED, Petrie EC, Thompson CE, Peskind ER (2000). The alpha1-adrenergic antagonist prazosin ameliorates combat trauma nightmares in veterans with posttraumatic stress disorder: a report of 4 cases. J Clin Psychiatry 61:129-133.

Raskind MA, Peskind ER, Hoff DJ, Hart KL, Holmes HA, Warren D, Shofer J, O'Connell J, Taylor F, Gross C, Rohde K, McFall ME (2007). A parallel group placebo controlled study of prazosin for trauma nightmares and sleep disturbance in combat veterans with post-traumatic stress disorder. Biol Psychiatry 61:928-934.

Raskind MA, Peskind ER, Kanter ED, Petrie EC, Radant A, Thompson CE, Dobie DJ, Hoff D, Rein RJ, Straits-Troster K, Thomas RG, McFall MM (2003). Reduction of nightmares and other PTSD symptoms in combat veterans by prazosin: a placebo-controlled study. Am J Psychiatry 160:371-373.

Rechtschaffen A, Kales A (1968). A manual of Standardized Terminology, Techniques and Scoring System for Sleep Stages of Human Subjects. Washington DC: US Government Printing Office.

Saper CB, Scammell TE, Lu J (2005). Hypothalamic regulation of sleep and circadian rhythms. Nature 437:1257-1263.

Smith SS, Dingwall K, Jorgenson G, Douglas J (2006). Associations between the use of common medications and sleep architecture in patients with untreated obstructive sleep apnea. J Clin Sleep Med 2:156-162.

Southwick SM, Bremner JD, Rasmusson A, Morgan CA, III, Arnsten A, Charney DS (1999). Role of norepinephrine in the pathophysiology and treatment of posttraumatic stress disorder. Biol Psychiatry 46:1192-1204.

Spitzer RL, Williams JB, Gibbon M, First MB (1992). The Structured Clinical Interview for DSM-III-R (SCID). I: History, rationale, and description. Arch Gen Psychiatry 49:624-629.

Taylor FB, Martin P, Thompson C, Williams J, Mellman TA, Gross C, Peskind ER, Raskind MA (2008). Prazosin effects on objective sleep measures and clinical symptoms in civilian trauma posttraumatic stress disorder: a placebo-controlled study. Biol Psychiatry 63:629-632.

van Liempt S, Vermetten E, Geuze E, Westenberg HG (2006). Pharmacotherapy for disordered sleep in post-traumatic stress disorder: a systematic review. Int Clin Psychopharmacol 21:193-202.

Chapter 8

Summary of major findings and discussion

SUMMARY OF MAJOR FINDINGS

In the introduction to this dissertation the relationship between disturbed sleep and post-traumatic stress disorder (PTSD) was addressed, as well as the importance of more research on the neurobiology of sleep in patients with PTSD. It was hypothesized that persistently disturbed sleep may be related to therapy-resistance and have an influence on daytime complaints.

Chapter 2 shows that nightmares, but not insomnia, are associated with an increased risk of developing PTSD symptoms after military deployment. This has been studied in a sample of 453 military service members who were deployed to Afghanistan. Screening for sleep disturbances and nightmares before deployment may contribute to early identification of those at risk of developing PTSD symptoms.

Chapter 3 shows that disturbed sleep due to obstructive apneas may exert a negative effect on PTSD symptoms as PTSD patients with obstructive sleep apnea (OSA) syndrome have more severe PTSD complaints. OSA is not more common in PTSD compared to trauma controls (TCs) and healthy controls (HCs).

Chapter 4 shows that sleep is more fragmented and heart rate is increased in PTSD compared to TCs and HCs. In this study polysomnography is combined with 20-minute-blood sampling. Plasma ACTH, cortisol, and melatonin concentrations are not significantly altered in PTSD, while a trend is seen for lower cortisol levels in the first half of the night. ACTH is positively related to the number of awakenings. Furthermore, ACTH is inversely related to the amount of slow wave sleep (SWS). PTSD patients exhibit a significantly decreased cortisol: ACTH ratio (CORT:ACTH) upon awakening compared to TCs. TCs demonstrate an increased CORT: ACTH during the night compared to both PTSD patients and HCs, suggesting an increased response of the adrenals upon ACTH stimulation under baseline conditions in veterans without lifetime psychiatric disorders.

Chapter 5 shows that PTSD patients have lower growth hormone levels during the night. Fragmented sleep is inversely related to growth hormone secretion. Noticeably, overnight memory consolidation is inversely related to awakenings. A positive relationship between memory recall and growth hormone secretion has been observed.

Chapter 6 comprises a systematic review, showing that pharmacotherapeutic options for sleep disturbances in PTSD have not been extensively studied with randomized controlled trials. So far, best results have been described in small RCTs investigating drugs with α_1 antagonistic properties.

In Chapter 7, treatment of sleep symptoms in PTSD patients with prazosin (n=6), a α_1-andrenoceptor antagonist, has no effect on the number of awakenings or other polysomnographic parameters and sleep diary measurements compared with placebo (n=6) in a small randomized controlled trial. Significantly more side effects occur after prazosin treatment.

REPERCUSSIONS OF DISTURBED SLEEP

Nightmares before military deployment, i.e. a period of increased risk of trauma exposure, increase the risk for PTSD symptom development in response to deployment. Furthermore, sleep apnea has been related to higher PTSD scores in PTSD patients. These observations indicate that disturbed sleep may influence PTSD symptomatology. Evidence from previous studies show that sleep positively affected generalization of extinction memory in healthy humans (Pace-Schott et al., 2009; Spoormaker et al., 2010; Spoormaker et al., 2011). This indicates that disturbed sleep may directly contribute to PTSD development by means of disrupting the beneficial process of sleep with regard to fear extinction. Nightmares predominantly occur during REM sleep (Nielsen and Levin, 2007), while they may also occur during NREM sleep in PTSD patients (Hefez et al., 1987). REM sleep in particular seems to have an effect on fear extinction (Spoormaker et al., 2011; Spoormaker et al., 2010). The observed relationship between nightmares and the risk of developing PTSD symptoms may suggest that disturbed REM sleep is a risk factor for PTSD.

On the other hand, nightmares have been associated with increased noradrenaline levels (Raskind et al., 2007). Furthermore, noradrenergic activity may be involved in PTSD development (Southwick et al., 1999). Thus, alternatively, the development of PTSD and the occurrence of nightmares may be epiphenomena, both induced by increased noradrenaline levels, and may not be causally linked. Our results suggest nightmares are a trait, making an individual more vulnerable to developing PTSD symptoms. Our study also shows that insomnia symptoms fail to predict PTSD symptoms at 6 months post deployment when pre-deployment mood and anxiety complaints are taken into account. These results contrast with previous studies in which a positive association was observed between the development of PTSD and insomnia symptoms before or directly after trauma exposure (Bryant et al., 2010; Koren et al., 2002; Mellman et al., 1995a). This may be explained by the fact that these studies did not correct for mood and anxiety complaints. The relationship between insomnia symptoms, and mood and anxiety complaints is complex and may be bidirectional: insomnia symptoms may contribute to mood and anxiety complaints, and – vice versa - insomnia symptoms may be moderated by mood or anxiety complaints (Abad and Guilleminault, 2005). Therefore, it is possible that mood and anxiety due to insomnia increase the risk for developing PTSD after trauma, and are therefore mediators in the relation between insomnia symptoms and PTSD development. Alternatively, mood and anxiety complaints may be confounding factors, causing both insomnia symptoms complaints and PTSD development. In the current design we could not differentiate whether mood and anxiety complaints are confounding factors or mediators in the relation between insomnia symptoms and PTSD development.

Obstructive sleep apnea Syndrome (OSAS) was not more prevalent in PTSD compared to trauma controls and healthy controls, which is in contrast with previous uncontrolled studies that suggested indices between 60 and 90% in PTSD (Krakow et al., 2004; Krakow et al., 2006; Krakow et al., 2000a; Krakow et al., 2002; Yesavage et al., 2010). In these studies screening instruments for detecting OSA may have been more sensitive than in our study, especially since some studies defined a cut-off of 5 events per hour. Another explanation for the high incidence of OSA in some previous studies is that the usage of benzodiazepines was not discontinued before sleep recordings, which increases the occurrence of OSA (Dolly and Block, 1982). In our study, participants with regular benzodiazepine usage were excluded, and participants with habitual benzodiazepine usage refrained from sleep medication in the sleep laboratory. Lastly, our study group consisted of middle-aged veterans, while other studies included either female PTSD patients, or elderly veterans with PTSD. The incidence of OSA may be different in other populations with PTSD. As none of the previous studies included a control group, it cannot be concluded that the incidence of OSA is elevated in PTSD. Our study underlines the importance of controlled studies to determine whether OSA is more prevalent in PTSD than in matched controls.

Nonetheless, we did observe a positive correlation between the apnea-hypopnea index per hour and more severe PTSD. Possibly, PSTD is a risk factor for OSAS, leading to a higher incidence of OSAS in severe PTSD. Alternatively, PTSD patients who happen to suffer from OSA as well, may experience symptom increases due to disturbed sleep. One uncontrolled studies has suggested a positive effect on PTSD symptoms after treating OSAS with continuous positive airway pressure (CPAP) (Krakow et al., 2000b). Possibly, when sleep is important for recovery, and is compromised by arousals due to obstructive events, OSAS may intervene with response to treatment. It would be advisable to screen for OSAS in case of snoring and other indicators of OSAS, especially in therapy resistant patients.

In this dissertation we also found a putative working mechanism for how sleep disturbances may influence daytime complaints. We found that growth hormone (GH) levels during the night were decreased in PTSD patients, compared to healthy controls. Reduced GH secretion correlated with awakenings during the night. A regression analyses with delayed recall as dependent showed that both sleep fragmentation and GH secretion were significant predictors for memory retention of a declarative memory task. This indicated that sleep-dependent memory consolidation is disturbed in PTSD due to decreased nocturnal GH secretion and more interrupted sleep. More research is warranted to confirm these novel findings.

Growth hormone receptors are present in the hippocampus (Lai et al., 1991). It is suggested that growth hormone stimulates neuroplasticity in the hippocampus (Kim et al., 2010). A recent functional MRI study in healthy volunteers showed decreased hippocam-

pus activation and decreased performance on a memory task after a night of experimentally induced sleep fragmentation (Van Der Werf et al., 2009). Thus, sleep fragmentation can have an effect on hippocampal function. In patients with PTSD also structural and functional changes in the hippocampus have been reported (Bremner, 2006). Interestingly, insomnia complaints in PTSD are inversely related to volume on the CA3 area of the hippocampus (Neylan et al., 2010). Decreased GH secretion may hypothetically be related to decreased hippocampus activity and possibly also hippocampal volume in PTSD. The effect of GH on neurons of the hippocampus during sleep deprivation has been shown in a recent preclinical study (Kim et al., 2010). This study shows that in the absence of GH during sleep deprivation N-methyl-D-aspartate (NMDA) receptor mediated synaptic currents decreased in hippocampal neurons. Moreover, NMDA receptor loss was observed, as was a decline in long term potentiation. These processes normalized when GH injections were administered during sleep deprivation. Reduced growth hormone secretion may possibly be related to decreased hippocampal functioning in PTSD. The relationship between treatment response to a SSRI and neurogenesis in the hippocampus was reported in a preclinical study (Santarelli et al., 2003). In combat-related PTSD, a relationship between treatment response and hippocampal volume has also been suggested (Vermetten et al., 2003). Future research should indicate whether GH secretion is related to hippocampus volume or functioning in PTSD. Furthermore, the relationship between hippocampal volume, neurogenesis and treatment-response remains to be elucidated.

CHARACTERISTICS OF SLEEP DISTURBANCES IN PTSD

Sleep in PTSD patients was characterized by more awakenings compared to TCs and HCs. Increased awakenings was also reported by earlier studies (Breslau et al., 2004; Habukawa et al., 2007; Mellman et al., 1995b). In agreement with a meta-analysis of polysomnographic (PSG) studies in PTSD total sleep time (TST) in our study was unchanged, as was Rapid Eye Movement sleep (REMS) (Kobayashi et al., 2007). Our study did not demonstrate reduced slow wave sleep (SWS), which was decreased according the meta-analysis on PSG studies in PTSD (Kobayashi et al., 2007).

In our study, cortisol levels tended to be lower during the first half of the night in patients with PTSD. ACTH levels were not elevated, which may have been due to the sample sizes (effect size $f = 0.42$, required total sample 58). Both cortisol and ACTH levels were correlated with SWS, while in a regression analysis only ACTH was an independent predictor for SWS. Interestingly, ACTH levels also correlated positively with the number of awakenings. ACTH secretion is stimulated by CRH activity. The correlation between ACTH and both SWS and awakenings may therefore be indicative for CRH being involved in waking

and SWS regulation in PTSD. CRH is known to inhibit SWS and increase waking during sleep (Steiger, 2007). It was previously observed that CRH is increased in cerebrospinal fluid and plasma in PTSD patients (Baker et al., 1999; De Kloet et al., 2008). Furthermore, in a meta-analysis on polysomnographic studies, SWS was decreased in PTSD. Further explorations of CRH in sleep complaints in PTSD may in time contribute to the development of novel treatment strategies.

We also hypothesized increased activity of the locus coeruleus (LC) in PTSD-related sleep fragmentation. The LC is a nucleus in the pons where noradrenergic neuronal cell bodies are located, which have projections throughout the brain. Also the sympathetic nervous system (SNS) is innervated by the LC. Noradrenergic activity stimulates waking by stimulating the ascending reticular arousal system (ARAS) and inhibiting sleep-promoting ventrolateral preoptic nucleus (VLPO) activity (Saper et al., 2005). Noradrenaline is therefore one of the key-players in the so-called "sleep/wake switch", a system involving the ARAS, VLPO and related neurotransmitters (Saper et al., 2005). We did indeed find increased heart rate in PTSD patients in comparison with TCs and HCs, which is in accordance with previous work and indicative for increased sympathetic activity (Muraoka et al., 1998;Woodward et al., 2009). We did not find any relationships between heart rate and awakenings in PTSD, nor in the combined sample. This may have been due to the small sample size; only eight PTSD patients could enter the analyses after excluding those with cardiovascular medication, which was used by a relative large portion of the PTSD patients (3/13). Recent research has indeed shown increased cardiovascular risk in PTSD (Boscarino, 2012). Possibly, the elevated SNS activity during the night may increase the risk for developing cardiovascular complications in PTSD.

The involvement of noradrenaline in PTSD related sleep disturbances was further supported by randomized controlled trials (RCTs) that demonstrate a positive effect of prazosin, an α_1-adrenoceptor blocker, on nightmares and insomnia in PTSD (Germain et al., 2011;Raskind et al., 2007;Raskind et al., 2003;Taylor et al., 2008).

In summary, heart rates were significantly higher during sleep in PTSD patients, indicating increased SNS activity. Furthermore, we found that awakenings were increased in PTSD. Interstingly, awakenings were positively related to ACTH secretion. In addition, perceived sleep quality was inversely related to the number of awakenings. Therefore, it is advised to calculate the number of awakenings in future PSG studies, as this seems to be a robust alteration in objective sleep quality in PTSD.

TREATMENT OF SLEEP DISTURBANCES IN PTSD

Insomnia and nightmares are frequently residual complaints in PTSD after successful psychotherapy with cognitive behaviour therapy (Zayfert and DeViva, 2004), or pharmacotherapy with SSRIs (Davidson et al., 2001). In Chapter 5 we systematically reviewed studies that were published before 2006, investigating pharmacotherapeutic options for sleep disturbances in PTSD. Our review shows that even though benzodiazepines are the most widely prescribed sleep medication, their effects on sleep disturbances in PTSD have not been extensively studied by RCTs. Only two small RCTs (n=6, n=22), with short treatment periods of 1 week, have been conducted (Mellman et al., 2002; Cates et al., 2004). Several small randomized controlled trials (RCT) have indicated that prazosin is superior to placebo in improving subjective sleep in PTSD (Germain et al., 2011; Raskind et al., 2003; Raskind et al., 2007; Taylor et al., 2008). Also, RCTs have shown the efficacy of the atypical antipsychotics olanzapine and risperidone as add-on therapy alongside a selective serotonin reuptake inhibitor (SSRI) (Rothbaum et al., 2008; Stein et al., 2002). Prazosin and quetiapine, another atypical antipsychotic, were similar in their short term effects (Byers et al., 2010). However, quetiapine more often led to discontinuation due to adverse side effects. One RCT showed that guanfacine, an $\alpha_2$adrenoceptor agonist, was not effective in treating nightmares and insomnia (Neylan et al., 2006).

Objective measurements of sleep were employed in two RCTs. One RCT investigating the effect of prazosin estimated REM sleep and total sleep time (TST) with an eye tracker (Taylor et al., 2008). This study suggested increased TST and REM sleep after prazosin treatment. Another RCT with three groups (prazosin, placebo and a cognitive behaviour therapy) measured polysomnography (PSG), the golden standard for measuring sleep architecture (Germain et al., 2011). In this study no differences over time were seen between groups on all PSG parameters and most subjective sleep measures. Only the number of reported nightmares was reduced in the active treatment groups in comparison with placebo. We also measured PSG and sleep questionnaires in a small RCT on the effect of prazosin, as has been described in chapter 6. Our study was in agreement with the observations from the study by Germain et al (2011) that PSG parameters, Pittsburgh sleep quality index (PSQI) scores and sleep diary measurements did not differ between groups. We also measured the number of awakenings pre and post treatment, but did not find a significant difference. This may have been due to the power of our study.

A meta-analysis including all RCTs on prazosin using PSG should be performed to further analyze the effect of prazosin on objective sleep quality in PTSD. However, PSG may not be a suitable method for detecting the underlying mechanism of nightmares in PTSD. Alternative methods may further elucidate the suggested effect of blocking α_1-adrenoceptor activity on nightmares. For instance, functional MRI with combined EEG

during sleep may demonstrate decreased activity of limbic brain structures during (REM) sleep in relation to a reduction in nightmare complaints.

In summary, prazosin and atypical antipsychotics are effective in the treatment of PTSD related sleep complaints. However, prazosin does not increase the risk for a metabolic syndrome and has therefore a more favourable side effect profile. One RCT showed that sleep focused behavioural therapy was as effective as prazosin on both insomnia and nightmares. Imaginary rehearsal therapy (IRT) is also effective for those suffering from nightmares, but cannot be applied in those without dream content recall. Prazosin treatment, imaginary rehearsal therapy (IRT) and sleep focused cognitive behavior therapy (CBT) are all effective for treating sleep disturbances (Germain et al., 2011; Krakow et al., 2001). Unfortunately, prazosin is no longer available in the Netherlands. Distribution has been stopped because prazosin was not regularly prescribed to patients suffering from hypertension and benign prostate hypertrophy. Alternatively, quetiapine, or the selective α_1-adrenoceptor antagonists doxazosin and alfuzosine, may be useful. However, RCTs are warranted to assess the efficacy of doxazosin and alfuzosine in PTSD-related sleep disturbances. Also sleep focused psychotherapy may not always be available to PTSD patients, as not all psychologists and psychiatrists are familiar with these interventions. The choice of treatment depends on the preference of the patient and availability of therapeutic options.

THE HPA-AXIS AND ADAPTATION AFTER TRAUMA EXPOSURE

Changes in the functioning of the hypothalamo-pituitary-adrenal (HPA) axis have repeatedly been associated PTSD, for a review see de Kloet et al (2006). Generally, increased responsivety to dexamethasone has been reported . Also, increased CRH levels have been demonstrated in cerebrospinal fluid and plasma (Baker et al., 1999; de Kloet et al., 2008). Results on peripheral cortisol values are inconsistent. Most studies report low or normal cortisol concentrations compared with controls (Klaassens et al., 2011; Meewisse et al., 2007). A previous study suggests that HPA-axis alterations after traumatic stress are also related to trauma exposure, and not merely with PTSD symptoms (de Kloet et al., 2007a; de Kloet et al., 2007b). We therefore included two control groups in our study; one group that comprised veterans without lifetime psychiatric disorder, and one control group of non-deployed -or otherwise traumatized- individuals. ACTH values in our study did not differ at night or in the morning between the three groups. Cortisol levels tended to be lower in PTSD during the first half of the night. However, significant group differences in the ratio CORT: ACTH were demonstrated. The CORT:ACTH ratio reflects the responsiveness of the adrenals upon ACTH stimulation. It appeared to be a sensitive measure for changes in HPA axis activity. It is advisable to test not only plasma cortisol in future

research, but also, if possible, to measure ACTH levels in order to calculate the ratio of CORT: ACTH.

In our small study, with a limited power to detect significant differences, we were able to demonstrated group differences in ratio CORT: ACTH. Firstly, an increased CORT: ACTH ratio was observed in trauma controls (TCs) during the night compared with both PSTD patients as healthy controls (HCs). This alteration in TCs compared with both PTSD patients and HCs was first described by Golier et al (2007) in Gulf War veterans (Golier et al., 2007). Additionally, in PTSD patients a decreased CORT: ACTH ratio was observed upon awakening compared with TCs. These observations may be ascribed to the responsiveness of the adrenals upon ACTH stimulation. Hyporesponsive adrenals to ACTH may underlie the decreased ratio in PTSD patients. In contrast, in TCs higher responsive adrenals to ACTH may explain the observed difference. We postulate that adaptation to trauma exposure leads to more responsive adrenals to ACTH, while a reduced responsiveness of the adrenals upon ACTH stimulation is seen in those who do not accomplish adaptation. This may implicate insufficient cortisol levels to induce an adequate inhibiting feedback response to HPA-axis activity in PTSD. In contrast, in TCs higher cortisol secretion may induce a more rapid normalization of a stress response, which may be reflected by "resilience" and adaptation.

Noticeably, glucocorticoid receptors (GR) are hyperresponsive in PTSD patients, who demonstrate exaggerated responses to dexamethasone administration (de Kloet et al., 2007b; Rohleder et al., 2004; Yehuda et al., 2004). A higher number of GR has also been reported before exposure to potentially traumatic events in service member who developed PTSD symptoms post deployment (van Zuiden et al., 2011). It is unknown whether in PTSD the reduced response of the adrenals to ACTH stimulation and elevated GR number and responsivety may be related. Presumably, low responsiveness of the adrenals to ACTH may imply relatively lower cortisol levels upon stimulation, and, subsequently, an up-regulation of GR receptors as a compensatory mechanism. One study also found elevated sensitivity of the GR receptor in TCs compared with healthy controls (de Kloet et al., 2007b). When cortisol secretion is increased in TCs after ACTH stimulation, and GR receptor enhancement takes place after trauma, the HPA-axis may be even further sensitized for the negative feedback response of cortisol in TCs.

Future research investigating stress and posttraumatic symptoms should focus on the dynamic characteristics of the HPA-axis, with for instance calculating cortisol: ACTH ratio's. In addition, sensitivity and up-regulation of receptors involved in the HPA-axis should be further explored.

CONCLUDING REMARKS AND RECOMMENDATIONS

Sleep has been studied with polysomnography (PSG) in the early days of PTSD research. However, the lack of objective findings in PSG studies urged the need to develop alternatives for measuring sleep in PTSD. This dissertation shows that sleep in PTSD is characterized by frequent awakenings at night, according to polysomnographic registrations. The number of awakenings in PTSD is associated with HPA-axis functioning, subjectively perceived sleep, and, interestingly, growth hormone secretion and memory consolidation. Furthermore, this dissertation supports the idea that disturbed sleep exerts a negative effect on PTSD symptomatology. Firstly, nightmares before military deployment predicted PTSD symptom development, independently early life trauma, mood and anxiety symptoms. Secondly, a positive correlation was observed between PTSD severity and the number of sleep apneas, which may indicate an increase in PTSD symptoms when sleep quality is compromised by external factors. Our results suggest that disturbed sleep increases the risk for PTSD. PTSD, in turn leads to increased sleep fragmentation, decreased growth hormone secretion and frequent nightmares, which may again compromise fear extinction, synaptic plasticity and recovery. This suggests that disturbed sleep is a precipitating and perpetuating factor in PTSD symptomatology, possibly creating a perpetual circle.

Longitudinal studies are warranted to investigate whether the differences in sleep fragmentation, growth hormone secretion and increased heart rate during sleep, found in our cross sectional studies, are "trait or state" phenomena. Additionally, more research is needed to further investigate the effects of sleep disruptions on fear extinction memory consolidation. New approaches are advised, such as combined functional MRI/ EEG studies to elucidate brain activation patterns and functional connectivity during sleep. More studies investigating sleep-dependent neuroplasticity and growth factors during sleep are also needed to further explore whether sleep disturbances affect biological underpinnings of psychiatric disease and recovery through the effect of sleep on synaptic plasticity.

REFERENCE LIST

Abad VC, Guilleminault C (2005). Sleep and psychiatry. Dialogues Clin Neurosci 7:291-303.

Baker DG, West SA, Nicholson WE, Ekhator NN, Kasckow JW, Hill KK, Bruce AB, Orth DN, Geracioti TD, Jr. (1999). Serial CSF corticotropin-releasing hormone levels and adrenocortical activity in combat veterans with posttraumatic stress disorder. Am J Psychiatry 156:585-588.

Boscarino JA (2012). PTSD is a risk factor for cardiovascular disease: Time for increased screening and clinical intervention. Prev Med.

Bremner JD (2006). Stress and brain atrophy. CNS Neurol Disord Drug Targets 5:503-512.

Breslau N, Roth T, Burduvali E, Kapke A, Schultz L, Roehrs T (2004). Sleep in lifetime posttraumatic stress disorder: a community-based polysomnographic study. Arch Gen Psychiatry 61:508-516.

Bryant RA, Creamer M, O'Donnell M, Silove D, McFarlane AC (2010). Sleep disturbance immediately prior to trauma predicts subsequent psychiatric disorder. Sleep 33:69-74.

Byers MG, Allison KM, Wendel CS, Lee JK (2010). Prazosin versus quetiapine for nighttime post-traumatic stress disorder symptoms in veterans: an assessment of long-term comparative effectiveness and safety. J Clin Psychopharmacol 30:225-229.

Cates ME, Bishop MH, Davis LL, Lowe JS, Woolley TW (2004). Clonazepam for treatment of sleep disturbances associated with combat-related posttraumatic stress disorder. Ann Pharmacother 38:1395-1399.

Davidson JR, Rothbaum BO, van der Kolk BA, Sikes CR, Farfel GM (2001). Multicenter, double-blind comparison of sertraline and placebo in the treatment of posttraumatic stress disorder. Arch Gen Psychiatry 58:485-492.

de Kloet CS, Vermetten E, Geuze E, Kavelaars A, Heijnen CJ, Westenberg HG (2006). Assessment of HPA-axis function in posttraumatic stress disorder: pharmacological and non-pharmacological challenge tests, a review. J Psychiatr Res 40:550-567.

de Kloet CS, Vermetten E, Bikker A, Meulman E, Geuze E, Kavelaars A, Westenberg HG, Heijnen CJ (2007a). Leukocyte glucocorticoid receptor expression and immunoregulation in veterans with and without post-traumatic stress disorder. Mol Psychiatry 12:443-453.

de Kloet CS, Vermetten E, Heijnen CJ, Geuze E, Lentjes EG, Westenberg HG (2007b). Enhanced cortisol suppression in response to dexamethasone administration in traumatized veterans with and without posttraumatic stress disorder. Psychoneuroendocrinology 32:215-226.

de Kloet CS, Vermetten E, Geuze E, Lentjes EG, Heijnen CJ, Stalla GK, Westenberg HG (2008). Elevated plasma corticotrophin-releasing hormone levels in veterans with posttraumatic stress disorder. Prog Brain Res 167:287-291.

Dolly FR, Block AJ (1982). Effect of flurazepam on sleep-disordered breathing and nocturnal oxygen desaturation in asymptomatic subjects. Am J Med 73:239-243.

Germain A, Richardson R, Moul DE, Mammen O, Haas G, Forman SD, Rode N, Begley A, Nofzinger EA (2011). Placebo-controlled comparison of prazosin and cognitive-behavior treatments for sleep disturbances in US Military Veterans. J Psychosom Res.

Golier JA, Schmeidler J, Legge J, Yehuda R (2007). Twenty-four hour plasma cortisol and adrenocorticotropic hormone in Gulf War veterans: relationships to posttraumatic stress disorder and health symptoms. Biol Psychiatry 62:1175-1178.

Habukawa M, Uchimura N, Maeda M, Kotorii N, Maeda H (2007). Sleep findings in young adult patients with posttraumatic stress disorder. Biol Psychiatry 62:1179-1182.

Hefez A, Metz L, Lavie P (1987). Long-term effects of extreme situational stress on sleep and dreaming. Am J Psychiatry 144:344-347.

Kim E, Grover LM, Bertolotti D, Green TL (2010). Growth hormone rescues hippocampal synaptic function after sleep deprivation. Am J Physiol Regul Integr Comp Physiol 298:R1588-R1596.

Klaassens ER, Giltay EJ, Cuijpers P, van VT, Zitman FG (2011). Adulthood trauma and HPA-axis functioning in healthy subjects and PTSD patients: A meta-analysis. Psychoneuroendocrinology.

Kobayashi I, Boarts JM, Delahanty DL (2007). Polysomnographically measured sleep abnormalities in PTSD: a meta-analytic review. Psychophysiology 44:660-669.

Koren D, Arnon I, Lavie P, Klein E (2002). Sleep complaints as early predictors of posttraumatic stress disorder: a 1-year prospective study of injured survivors of motor vehicle accidents. Am J Psychiatry 159:855-857.

Krakow B, Germain A, Tandberg D, Koss M, Schrader R, Hollifield M, Cheng D, Edmond T (2000a). Sleep breathing and sleep movement disorders masquerading as insomnia in sexual-assault survivors. Compr Psychiatry 41:49-56.

Krakow B, Haynes PL, Warner TD, Santana E, Melendrez D, Johnston L, Hollifield M, Sisley BN, Koss M, Shafer L (2004). Nightmares, insomnia, and sleep-disordered breathing in fire evacuees seeking treatment for posttraumatic sleep disturbance. J Trauma Stress 17:257-268.

Krakow B, Hollifield M, Johnston L, Koss M, Schrader R, Warner TD, Tandberg D, Lauriello J, McBride L, Cutchen L, Cheng D, Emmons S, Germain A, Melendrez D, Sandoval D, Prince H (2001). Imagery rehearsal therapy for chronic nightmares in sexual assault survivors with posttraumatic stress disorder: a randomized controlled trial. JAMA 286:537-545.

Krakow B, Lowry C, Germain A, Gaddy L, Hollifield M, Koss M, Tandberg D, Johnston L, Melendrez D (2000b). A retrospective study on improvements in nightmares and post-traumatic stress disorder following treatment for co-morbid sleep-disordered breathing. J Psychosom Res 49:291-298.

Krakow B, Melendrez D, Warner TD, Clark JO, Sisley BN, Dorin R, Harper RM, Leahigh LK, Lee SA, Sklar D, Hollifield M (2006). Signs and symptoms of sleep-disordered breathing in trauma survivors: a matched comparison with classic sleep apnea patients. J Nerv Ment Dis 194:433-439.

Krakow B, Melendrez D, Warner TD, Dorin R, Harper R, Hollifield M (2002). To breathe, perchance to sleep: sleep-disordered breathing and chronic insomnia among trauma survivors. Sleep Breath 6:189-202.

Lai ZN, Emtner M, Roos P, Nyberg F (1991). Characterization of putative growth hormone receptors in human choroid plexus. Brain Res 546:222-226.

Mahan AL, Ressler KJ (2012). Fear conditioning, synaptic plasticity and the amygdala: implications for posttraumatic stress disorder. Trends Neurosci 35:24-35.

Meewisse ML, Reitsma JB, de Vries GJ, Gersons BP, Olff M (2007). Cortisol and post-traumatic stress disorder in adults: systematic review and meta-analysis. Br J Psychiatry 191:387-392.

Mellman TA, Bustamante V, David D, Fins AI (2002). Hypnotic medication in the aftermath of trauma. J Clin Psychiatry 63:1183-1184.

Mellman TA, David D, Kulick-Bell R, Hebding J, Nolan B (1995a). Sleep disturbance and its relationship to psychiatric morbidity after Hurricane Andrew. Am J Psychiatry 152:1659-1663.

Mellman TA, Kulick-Bell R, Ashlock LE, Nolan B (1995b). Sleep events among veterans with combat-related posttraumatic stress disorder. Am J Psychiatry 152:110-115.

Muraoka MY, Carlson JG, Chemtob CM (1998). Twenty-four-hour ambulatory blood pressure and heart rate monitoring in combat-related posttraumatic stress disorder. J Trauma Stress 11:473-484.

Neylan TC, Lenoci M, Samuelson KW, Metzler TJ, Henn-Haase C, Hierholzer RW, Lindley SE, Otte C, Schoenfeld FB, Yesavage JA, Marmar CR (2006). No improvement of posttraumatic stress disorder symptoms with guanfacine treatment. Am J Psychiatry 163:2186-2188.

Neylan TC, Mueller SG, Wang Z, Metzler TJ, Lenoci M, Truran D, Marmar CR, Weiner MW, Schuff N (2010). Insomnia severity is associated with a decreased volume of the CA3/dentate gyrus hippocampal subfield. Biol Psychiatry 68:494-496.

Nielsen T, Levin R (2007). Nightmares: a new neurocognitive model. Sleep Med Rev 11:295-310.

Pace-Schott EF, Milad MR, Orr SP, Rauch SL, Stickgold R, Pitman RK (2009). Sleep promotes generalization of extinction of conditioned fear. Sleep 32:19-26.

Raskind MA, Peskind ER, Kanter ED, Petrie EC, Radant A, Thompson CE, Dobie DJ, Hoff D, Rein RJ, Straits-Troster K, Thomas RG, McFall MM (2003). Reduction of nightmares and other PTSD symptoms in combat veterans by prazosin: a placebo-controlled study. Am J Psychiatry 160:371-373.

Raskind MA, Peskind ER, Hoff DJ, Hart KL, Holmes HA, Warren D, Shofer J, O'Connell J, Taylor F, Gross C, Rohde K, McFall ME (2007). A parallel group placebo controlled study of prazosin for trauma nightmares and sleep disturbance in combat veterans with post-traumatic stress disorder. Biol Psychiatry 61:928-934.

Rohleder N, Joksimovic L, Wolf JM, Kirschbaum C (2004). Hypocortisolism and increased glucocorticoid sensitivity of pro-Inflammatory cytokine production in Bosnian war refugees with posttraumatic stress disorder. Biol Psychiatry 55:745-751.

Rothbaum BO, Killeen TK, Davidson JR, Brady KT, Connor KM, Heekin MH (2008). Placebo-controlled trial of risperidone augmentation for selective serotonin reuptake inhibitor-resistant civilian posttraumatic stress disorder. J Clin Psychiatry 69:520-525.

Santarelli L, Saxe M, Gross C, Surget A, Battaglia F, Dulawa S, Weisstaub N, Lee J, Duman R, Arancio O, Belzung C, Hen R (2003). Requirement of hippocampal neurogenesis for the behavioral effects of antidepressants. Science 301:805-809.

Saper CB, Scammell TE, Lu J (2005). Hypothalamic regulation of sleep and circadian rhythms. Nature 437:1257-1263.

Southwick SM, Bremner JD, Rasmusson A, Morgan CA, III, Arnsten A, Charney DS (1999). Role of norepinephrine in the pathophysiology and treatment of posttraumatic stress disorder. Biol Psychiatry 46:1192-1204.

Spoormaker VI, Schroter MS, Andrade KC, Dresler M, Kiem SA, Goya-Maldonado R, Wetter TC, Holsboer F, Samann PG, Czisch M (2011). Effects of rapid eye movement sleep deprivation on fear extinction recall and prediction error signaling. Hum Brain Mapp.

Spoormaker VI, Sturm A, Andrade KC, Schroter MS, Goya-Maldonado R, Holsboer F, Wetter TC, Samann PG, Czisch M (2010). The neural correlates and temporal sequence of the relationship between shock exposure, disturbed sleep and impaired consolidation of fear extinction. J Psychiatr Res 44:1121-1128.

Steiger A (2007). Neurochemical regulation of sleep. J Psychiatr Res 41:537-552.

Stein MB, Kline NA, Matloff JL (2002). Adjunctive olanzapine for SSRI-resistant combat-related PTSD: a double-blind, placebo-controlled study. Am J Psychiatry 159:1777-1779.

Taylor FB, Martin P, Thompson C, Williams J, Mellman TA, Gross C, Peskind ER, Raskind MA (2008). Prazosin effects on objective sleep measures and clinical symptoms in civilian trauma posttraumatic stress disorder: a placebo-controlled study. Biol Psychiatry 63:629-632.

Troxel WM, Kupfer DJ, Reynolds CF, III, Frank E, Thase ME, Miewald JM, Buysse DJ (2011). Insomnia and objectively measured sleep disturbances predict treatment outcome in depressed patients treated with psychotherapy or psychotherapy-pharmacotherapy combinations. J Clin Psychiatry.

Van Der Werf YD, Altena E, Schoonheim MM, Sanz-Arigita EJ, Vis JC, De RW, Van Someren EJ (2009). Sleep benefits subsequent hippocampal functioning. Nat Neurosci 12:122-123.

van Zuiden M, Geuze E, Willemen HL, Vermetten E, Maas M, Heijnen CJ, Kavelaars A (2011). Pre-existing high glucocorticoid receptor number predicting development of posttraumatic stress symptoms after military deployment. Am J Psychiatry 168:89-96.

Vermetten E, Vythilingam M, Southwick SM, Charney DS, Bremner JD (2003). Long-term treatment with paroxetine increases verbal declarative memory and hippocampal volume in posttraumatic stress disorder. Biol Psychiatry 54:693-702.

Woodward SH, Arsenault NJ, Voelker K, Nguyen T, Lynch J, Skultety K, Mozer E, Leskin GA, Sheikh JI (2009). Autonomic activation during sleep in posttraumatic stress disorder and panic: a mattress actigraphic study. Biol Psychiatry 66:41-46.

Yehuda R, Halligan SL, Golier JA, Grossman R, Bierer LM (2004). Effects of trauma exposure on the cortisol response to dexamethasone administration in PTSD and major depressive disorder. Psychoneuroendocrinology 29:389-404.

Yesavage JA, Kinoshita LM, Kimball T, Zeitzer J, Friedman L, Noda A, David R, Hernandez B, Lee T, Cheng J, O'Hara R (2010). Sleep-Disordered Breathing in Vietnam Veterans with Posttraumatic Stress Disorder. Am J Geriatr Psychiatry.

Zayfert C, DeViva JC (2004). Residual insomnia following cognitive behavioral therapy for PTSD. J Trauma Stress 17:69-73.

Chapter 9

Nederlandse samenvatting

SLAAPSTOORNISSEN EN POSTTRAUMATISCHE STRESSSTOORNIS; EEN VICIEUZE CIRKEL?

INLEIDING

Slaap is voor ieder mens van vitaal belang. Een gezonde slaap draagt bij aan herstel en geheugenopslag. Nachtmerries en slapeloosheid komen bij patiënten met een posttraumatische stress stoornis veelvuldig voor. Deze slaapstoornissen leiden tot een hoge lijdensdruk. Bovendien is slaap mogelijk relevant voor herstel. Verstoorde slaap kan een remmend effect kan hebben op de vorming van nieuwe zenuwverbindingen en op het uitdoven van een angstreactie; de "herinnering" aan de uitdoving is afhankelijk van een goede slaapkwaliteit. En juist dit uitdoven van angstreacties blijkt bij patiënten met PTSS niet goed te verlopen. Er zijn redenen om aan te nemen dat een gebrekkige of onvoldoende uitdoving van angst samenhangt met de slaapstoornissen bij PTSS patiënten. De vraag is of slaapstoornissen niet alleen het gevolg van de PTSS zijn en of ze PTSS klachten wellicht ook in stand houden. Is verstoorde slaap misschien zelfs een risico factor voor PTSS? In dit geval zou er sprake kunnen zijn van een vicieuze cirkel, waarbij slaapverstoringen eerst bijdragen aan het ontstaan van PTSS, vervolgens door PTSS verder verstoord raken, en tenslotte de PTSS klachten in stand houden.

In dit proefschrift staan verschillende studies beschreven die een verband tussen slaapstoornissen en PTSS onderzochten. Ook werd de samenhang bestudeerd tussen slaapklachten en verschillende neurobiologische systemen tijdens slaap: het sympathische zenuwstelsel, groeihormoonsecretie, en de hypothalamus-hypofyse-bijnier as (HPA-as, welke bestaat uit de gerelateerde stresshormonen corticotropin-releasing hormone (CRF), adrenocorticotroop hormoon (ACTH) en cortisol. Tenslotte werden ook behandelmogelijkheden voor slaapklachten bij PTSS onderzocht.

HOOFDSTUK 2

De vraag of slaapstoornissen een risicofactor zijn voor het ontwikkelen van PTSS klachten na uitzending werd onderzocht in een prospectieve cohort studie. Bij 453 militairen werd voor uitzending naar Afghanistan gevraagd in hoeverre er sprake was van nachtmerries en slapeloosheid. Hiervoor werden twee vragenlijsten gebruikt: de Symptom Checklist -90 (SCL-90) en de Zelfinventarisatielijst voor PTSS klachten (ZIL). Bovendien werden potentieel traumatische ervaringen in de jeugd uitgevraagd met de Early Trauma Inventory (ETI). Depressieve klachten en angstklachten voorafgaande aan uitzending werden vastge-

steld met de SCL-90. Zes maanden na uitzending werden middels de ZIL eventuele PTSS symptomen vastgesteld.

Uit dit onderzoek bleek dat nachtmerries een significante voorspeller zijn voor PTSS klachten, onafhankelijk van depressie, angst, eerdere uitzendingen of trauma op de kinderleeftijd. Voor klachten van slapeloosheid gold echter dat de voorspellende waarde niet langer significant was na correctie voor depressie en angstklachten.

HOOFDSTUK 3

Of een obstructief slaapapneusyndroom regelmatig voorkomt bij PTSS patiënten en correleert met de ernst van PTSS klachten, werd onderzocht in hoofdstuk 3. Veteranen met PTSS (n=20), veteranen zonder PTSS (n=24) en controles zonder psychotrauma (n=17) namen deel aan dit onderzoek. Iedere proefpersoon werd 1 nacht onderworpen aan een slaaponderzoek met behulp van "polysomnografie", wat inhoud dat de slaapstructuur en de ademhaling tijdens de nacht geregistreerd werd. Bij de drie groepen kwamen slaapapneus evenveel voor. Er was geen relatie tussen het aantal slaapapneus en het optreden van nachtmerries. Wel werd er een positief verband gevonden tussen de ernst van PTSS klachten en het aantal slaapapneus.

Geconcludeerd werd dat verstoring van de slaap door externe factoren, zoals slaapapneus, mogelijk zou kunnen leiden tot een verergering van PTSS klachten.

HOOFDSTUK 4

In hoofdstuk 4 wordt het onderzoek naar verschillen in de slaapstructuur, de hartslag en plasma concentratie van cortisol, ACTH en melatonine tijdens de slaap tussen de drie groepen proefpersonen beschreven. Personen bij wie het slaapapneu syndroom werd geconstateerd, werden uitgesloten van dit onderzoek. De overige proefpersonen bestonden uit 13 veteranen met PTSS (PTSS), 17 veteranen zonder PTSS en 15 niet-veteranen zonder psychotrauma's. Deze proefpersonen ondergingen opnieuw een polysomnografie, dit keer voor twee nachten. Tijdens de laatste nacht werden er tegelijkertijd bloedafnamen gedaan vanuit een infuus, zonder participanten te storen in de slaap. Centraal stond de vraag of er tussen de groepen verschillen zijn in slaapstructuur, hartslag en concentratie van cortisol, ACTH en melatonine tijdens de slaap.

Bij PTSS patiënten bleek de slaap veel vaker onderbroken door korte periodes van waak, terwijl de totale slaap tijd niet afweek. Het aantal keer dat iemand wakker werd ('wekreacties') hing samen met de concentratie ACTH in de nacht. Ook de subjectieve indruk van de slaapdiepte hing samen met het aantal wekreacties, terwijl de hoeveelheid

diepe slaap bij veteranen met PTSS niet afwijkend was. De hartslag tijdens de slaap was hoger in de groep deelnemers met PTSS vergeleken de beide controlegroepen. Er werd geen correlatie gevonden tussen de hartslag en de slaapkwaliteit. De ratio cortisol/ACTH tijdens de nacht was bij veteranen zonder PTSS significant hoger dan bij de PTSS patiënten en ook hoger dan bij de controlegroep zonder psychotrauma. De ratio was ook verhoogd bij het ontwaken, echter dan alleen ten opzichte van PTSS patiënten.

Het aantal wekreacties blijkt een belangrijke afwijking tijdens de slaap van PTSS patienten, die zowel samenhangt met subjectieve beleving van slaap, als met activiteit van de HPA-as. Verder duidt de verhoogde hartslag op verhoogde sympathische activiteit tijdens slaap, mogelijk samenhangend met een verhoogde activiteit van de locus coeruleus.

HOOFDSTUK 5

Tijdens de slaapregistratie beschreven in het vorige hoofdstuk, werd tevens de groeihormoon concentratie in het bloed gemeten. De avond voor het slaaponderzoek werd de deelnemers in drie rondes 15 woorden geleerd. De ochtend na de slaapregistratie werd gevraagd hoeveel woorden zij nog kenden. Waar de plasma concentratie groeihormoon normaal piekt in het eerste trimester van de nacht, bleek de groeihormoon concentratie significant verlaagd bij PTSS patiënten. PTSS patiënten scoorden tevens slechter op de geheugentest in de ochtend. Er bleek een positief verband tussen geheugenopslag en de concentratie groeihormoon en een negatief verband tussen geheugen en het aantal wekreacties. Wekreacties en groeihormoonsecretie correleerden ook met elkaar, maar beiden hadden een onafhankelijk effect op de geheugenopslag.

HOOFDSTUK 6

Een literatuurstudie naar effectieve medicamenteuze behandelopties voor slapeloosheid en nachtmerries bij PTSS patiënten laat zien dat er verschillende open-label studies en case-reports zijn gepubliceerd met positieve resultaten voor verschillende antidepressiva en enkele antiepileptica. Kleine gecontroleerde studies suggereren dat het atypische antipsychoticum olanzapine en de adrenerge α_1-blokker prazosine werkzaam zijn bij de behandeling van slaapklachten bij patiënten met PTSS.

HOOFDSTUK 7

Om te onderzoeken of prazosine het aantal wekreacties kan laten afnemen, werd een dubbelblinde gecontroleerde studie uitgevoerd bij 14 veteranen, waarvan 7 deelnemers acht weken werden behandeld met prazosine en 7 deelnemers met placebo. Het was onbekend voor de deelnemers en de onderzoekers wie wat gebruikte. Deelnemers ondergingen twee nachten slaapregistratie in hun eigen huis voor de behandeling en in de laatste week van de behandeling. In iedere groep viel een deelnemer uit, in de prazosine groep wegens bijwerkingen, in de placebo groep wegens de klinische noodzaak om antidepressieve medicatie te wijzingen tijdens het onderzoek. Na behandeling met prazosine werd echter in deze studie geen verbetering van de slaapkwaliteit, noch een afname van het aantal wekreacties gezien ten opzichte van de placebogroep.

CONCLUSIE

Onze bevinding dat nachtmerries voor uitzending de kans op een PTSS verhogen sluit aan bij het idee van een causale relatie tussen slaapverstoringen en het ontstaan van PTSS. Ook de relatie tussen het aantal slaapapneus en de ernst van PTSS klachten kan passen bij een oorzakelijk verband tussen slaapverstoringen en PTSS symptomatologie. Verder laat dit proefschrift zien dat PTSS geassocieerd is met een sterk onderbroken slaap en verminderde groeihormoonsecretie tijdens de slaap. Verstoorde slaap zou de kans op een goed herstel van de PTSS klachten kunnen verhinderen, waardoor een vicieuze cirkel kan ontstaan. De opslag van declaratief geheugen was in onze studie verminderd bij PTSS. Het effect van de slaaponderbrekingen en verminderde groeihormoon secretie op de consolidatie van verschillende domeinen van het geheugen verdient meer aandacht in toekomstige studies bij PTSS, maar ook bij andere psychiatrische stoornissen die een chronisch beloop kunnen hebben. Bij PTSS is het met name interessant of de verwerking van emotioneel geheugen en de uitdoving van angstreacties afhangt van de slaapkwaliteit. Dit zou namelijk het neurobiologische mechanisme kunnen zijn van een "vicieuze cirkel". Waar de verstoorde slaap door veroorzaakt wordt blijft grotendeels onbekend. Dit proefschrift leverde aanwijzingen voor betrokkenheid van zowel de activiteit van de HPA-as activiteit als activiteit van het sympathische zenuwstelsel, en dan met name de centrale (α_1-adrenoceptor gemedieerde) noradrenerge activiteit. Of de HPA-as en sympathische zenuwstelsel inderdaad samenhangen met slaapkwaliteit bij PTSS en een bruikbaar focus vormen voor de behandeling zal toekomstig onderzoek moeten uitwijzen. Het is van groot belang dit verder uit te zoeken omdat gezonde slaap bij PTSS van essentieel belang zou kunnen zijn voor herstel.

Dankwoord

Dankwoord

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Publications

Publications

- van Liempt, S., Vermetten, E., Lentjes, E., Arends, J., & Westenberg, H. (2011). Decreased nocturnal growth hormone secretion and sleep fragmentation in combat-related posttraumatic stress disorder; potential predictors of impaired memory consolidation. *Psychoneuroendocrinology*, 36(9), 1361-9

- van Liempt, S., Westenberg, H., Arends, J., Vermetten, E (2011). Obstructive Sleep Apnea in Combat-related Post-traumatic Stress Disorder; a controlled polysomnography study. *European Journal of Psychotraumatology 2011; **2**: 8451-5*

- van Liempt S, Vermetten E, Geuze E, Westenberg HG (2006). Pharmacotherapy for disordered sleep in post-traumatic stress disorder: a systematic review. *Int Clin Psychopharmacol* 21:193-202

- Ariagno RL, van Liempt S, Mirmiran M (2006). Fewer spontaneous arousals during prone sleep in preterm infants at 1 and 3 months corrected age. *J Perinatol*. 26(5):306-12

- van Liempt S, Vermetten E, Geuze E, Westenberg H (2006). Pharmacotherapeutic treatment of nightmares and insomnia in posttraumatic stress disorder: an overview of the literature. *Ann N Y Acad Sci.* 1071:502-7. Review.

- van Liempt S, Vermetten E, de Groen JH, Westenberg HG (2007). Slaapafwijkingen bij posttraumatische stressstoornis; overzicht van onderzoeksbevindingen. *Tijdschr Psychiatr.* 2007;49(9):629-38. Review.

SUBMITTED

- van Liempt, S., van Zuiden, Z., Westenberg, H., Super, A., Vermetten, E. The impact of impaired sleep as predictor of PTSD symptoms in combat veterans; a prospective longitudinal cohort study *(submitted)*

- van Liempt, S., Arends, J., Cluitmans., P.J.M., Westenberg, H., Kahn, R.S., Vermetten, E. Sympathetic activity and hypothalamo-pituitary-adrenal axis activity during sleep in post-traumatic stress disorder; a study assessing polysomnography with simultaneous blood sampling *(submitted)*

Curriculum Vitae

Curriculum Vitae

Saskia van Liempt (1978) finished secondary school at the Montessori Lyceum Amsterdam in 1997, after which she started studying Medicine at the University of Amsterdam. In 2001 she studied the effect of body position on sleep and arousals in preterm infants during a scientific internship at Stanford University, California. In 2004 she obtained her medical degree after an internship in tropical medicine in Ndala, Tanzania. She started working at the Military Psychiatry, Utrecht, as a researcher and physician in 2004. From 2005 until 2006 she worked as a resident in sleep medicine at Kempenhaeghe, Heeze. In 2006 she started her dissertation on sleep disturbances in veterans with PTSD at the Military Mental Health Care under the supervision of prof. dr. H.G.M. Westenberg, prof. dr. J. Arends and dr. E. Vermetten. She started her residency in Psychiatry in 2010 at the University Medical Centre in Utrecht under supervision of dr. J. Wijkstra and prof. dr. R.S. Kahn. From 2011 prof. dr. R.S. Kahn also supervised the final phase of her dissertation. Saskia van Liempt lives in Utrecht with Onno Kwast and their daughter Rosa.