Fachbereich Erziehungswissenschaft und Psychologie
der Freien Universität Berlin

Neuropsychoendocrinological and Psychophysiological Aspects of Trauma Memory

– An examination with a trauma film paradigm –

Dissertation
zur Erlangung des akademischen Grades
Doktorin der Philosophie (Dr. phil.)

vorgelegt von
Dipl.-Psych.
Felicitas Rombold-Brühl

Berlin, 2017

Erstgutachterin (first supervisor):
Prof. Dr. rer.nat. Babette Renneberg

Zweitgutachter (second supervisor):
Prof. Dr. med. Stefan Röpke

Tag der Disputation (day of thesis defense):
08.03.2018

Dedicated to Anton Hanno Brühl

who was born during the last year of this dissertation

Acknowledgements

First of all I would like to give my warmest thanks to Prof. Dr. Stefan Röpke. I could have not wished for a better supervision. Thank you for the generous guidance from the beginning to the end, the invaluable support, and appreciative feedback. I am truly grateful to have worked with you and to have learned from your immense expertise!

Furthermore, I would like to express my gratitude to Prof. Dr. Babette Renneberg for supervising this dissertation, supporting me during the scholarship application process, and giving constructive feedback on the study designs, the manuscripts, and the dissertation theses.

Next, I wish to thank Prof. Dr. Christian Otte and Prof. Dr. Katja Wingenfeld for their involvement in the planning process of the studies, their critical feedback on the manuscripts, and the weekly 'stress and memory' team meetings. Without your help this theses would have not been possible. Additionally, I would like to thank Prof. Dr. Christian Otte for his support for my post-doc scientific career.

I further want to thank Prof. Dr. Christine Knaevelsrud, Prof. Dr. Nina Knoll, Dr. Sarah Schumacher, and Prof. Dr. Stephan Heinzel who kindly agreed to be part of the dissertation defense committee.

Further, I would like to thank the federal state of Berlin for awarding me the Elsa-Neumann-Stipendium.

Additionally, I thank all my colleagues at Charité Universitätsmedizin Berlin for the inspiring scientific conversations, the motivating smiles on the hallways, and the friendships that have grown during the time at the clinic.

Moreover, I would like to thank all my friends for their emotional support during the past years. Special thanks goes to my dear friend Julia Wysokowsky who worked with a trauma film paradigm before and supported me with her initial assistance during the planning process.

Many more people have to be thanked: Robert for cutting the movie material, Dr. Larissa Wolkenstein for her support during the grant application process, Jennifer Mumm for being my heart rate variability partner in crime, Anna Schmied, Linda Bruch, and Friederike Schwarzkopf for watching the trauma film with the participants many, many times as part of their master thesis, Dr. Julian

Hellmann for analyzing the saliva samples, PD Dr. Frank Zimmermann-Viehoff for incorporating me into the world of heart rate variability, Dr. Christina Anderson and Stephanie & Robert O'Brien for proof reading the many different versions of the manuscripts, and Dr. Anke Weidmann for passing on her knowledge on the design.

Finally, I would like to thank my family. I am deeply grateful to my parents Birgit Rombold and Andreas Rombold for their unconditional love and guidance throughout my life. Thank you for letting me unfold my personality and potentials in a secure environment. Also I want to thank my siblings Larissa and Fabian Rombold for the invaluable help with Anton while I was writing this dissertation and their emotional support.

Most importantly, I thank my husband Daniel Brühl for the extended walks with our baby, the support on this long educational way, the interest in my profession, and his love. Muchas gracias mi alma.

Table of contents

The stream of thought flows on; but most of its segments fall into the bottomless abyss of oblivion. Of some, no memory survives the instant of their passage. Of others, it is confined to a few moments, hours, or days. Others, again, leave vestiges which are indestructible, and by means of which they may be recalled as long as life endures. Can we explain these differences? (William James, 1890, p.643)

English Summary

Intrusive memories and memory impairments of key features of the trauma are symptoms of posttraumatic stress disorder (PTSD). The underlying biological mechanisms impacting on trauma related memories are not sufficiently understood. Stress hormones such as cortisol and noradrenaline are involved in emotional memory formation. However, little is known about neuroendocrinological influences during trauma on subsequent intrusive memories and recognition of trauma details. Further, first evidence suggests an association between lower baseline heart rate variability, an indicator of autonomic nervous system activity, and increased risk for PTSD. However, the association between heart rate variability and intrusive memories remains to be examined.

The aim of the presented dissertation is to extend the existing models on intrusion formation with regard to whether and how endocrinological and physiological aspects impact on intrusion formation and recognition of trauma details. Therefore, the following research questions are examined within an intrusion-inducing trauma film paradigm in healthy women:

(1) Does the activity of the noradrenergic system during a trauma film influence subsequent intrusive memories?
(2) Does the activity of the hypothalamus–pituitary–adrenal (HPA) axis during a trauma film influence subsequent intrusive memories?
(3) Is HRV at rest before a trauma film associated with subsequent intrusive memories?
(4) Does the activity of the noradrenergic system and the HPA axis during a trauma film influence subsequent memory for trauma film details?

The main results of this dissertation are as follows. First, pharmacologically increased noradrenergic activity during a trauma film led to a delayed decrease of the number and vividness of subsequent intrusions compared to pharmacologically decreased noradrenergic activity and placebo. Second, pharmacologically increased activity of the HPA axis during the trauma film did not

affect subsequent intrusive memories. Third, pharmacologically decreased noradrenergic activity during the trauma film led to significantly fewer correct peri-trauma film scene memories compared to placebo and, on a trend level, to pharmacologically increased noradrenergic activity. Fourth, lower baseline heart rate variability before the trauma film was associated with a delayed decrease of the number of intrusive memories.

To conclude, the conducted trauma film paradigm studies offered the opportunity to study mechanisms that are impossible to study during real life trauma. The findings of this dissertation extend models on intrusion formation by emphasizing and including endocrinological and physiological aspects. This contributes to a relevant growth of knowledge on trauma related memory and PTSD. The results of this dissertation project shed light on endocrinological and physiological aspects of intrusive memory formation and recognition of trauma details.

Deutsche Zusammenfassung (German Summary)

Intrusives Wiedererleben und Beeinträchtigungen des Erinnerungsvermögens an Hauptmerkmale des Traumas, sind typische Symptome der Posttraumatischen Belastungsstörung (PTBS). Welche Faktoren die Entstehung dieser Symptome beeinflussen, ist nicht hinreichend verstanden. Die Stresshormone Kortisol und Noradrenalin modulieren emotionale Gedächtnisbildung, jedoch ist wenig über ihren Einfluss während der Akquisition und Konsolidierung eines traumatischen Ereignisses auf nachfolgende Intrusionen und Erinnerungen an das Trauma bekannt. Zusätzlich zeigen erste Befunde einen Zusammenhang zwischen niedriger Herzratenvariabilität (HRV), einem Indikator für die Aktivität des autonomen Nervensystems, und einem erhöhten Risiko für PTSD. Der Zusammenhang zwischen HRV und Intrusionen ist jedoch noch unklar.

Gegenstand dieser Dissertation ist es bisherige Modelle zu traumabezogenen Erinnerungen zu erweitern, indem herausgefunden wird, ob und inwieweit neuroendokrinologische Faktoren oder physiologische Faktoren einen Einfluss auf die Intrusionsbildung und das Wiedererkennen von Traumadetails haben. Dafür werden folgende Hauptfragestellungen im Rahmen des Trauma-Film-Paradigmas mit gesunden Frauen untersucht:

(1) Beeinflusst die Aktivität des noradrenergen Systems während eines Traumafilmes nachfolgende intrusive Erinnerungen?
(2) Beeinflusst die Aktivität der Hypothalamus-Hypophysen-Nebennierenachse während eines Traumafilmes nachfolgende intrusive Erinnerungen?
(3) Ist die HRV vor einem Traumafilm mit intrusiven Erinnerungen nach diesem Traumafilm assoziiert?
(4) Beeinflusst die Aktivität des noradrenergen System und der HPA-Achse während eines Traumafilmes nachfolgende Erinnerungen an diesen Traumafilm?

Die Hauptbefunde dieser Dissertation sind wie folgt. Erstens, eine pharmakologische Aktivierung des noradrenergen Systems zum Zeitpunkt der

Akquisition und Konsolidierung des Traumafilmes führte zu einem späteren Abfall der Intrusionen im Vergleich zu einer pharmakologisch verminderten Aktivität des noradrenergen Systems und einer Placebobedingung. Zweitens, eine pharmakologisch erhöhte Aktivität der HPA-Achse zum Zeitpunkt der Akquisition und Konsolidierung des Traumafilmes hatte keinen Einfluss auf nachfolgende Intrusionen. Drittens, eine pharmakologisch verminderte Aktivität des noradrenergen Systems zum Zeitpunkt der Akquisition und Konsolidierung des Traumafilmes führte zu weniger richtigen peritraumatischen Filmerinnerungen im Vergleich zu Placebo. Viertens, eine niedrigere HRV vor dem Traumafilm war mit einer höheren Anzahl an Intrusionen nach dem Traumafilm assoziiert.

Die durchgeführten Trauma-Film-Paradigma Studien ermöglichten das experimentelle Untersuchen von Mechanismen, die traumabezogenen Erinnerungen zugrunde liegen und bei echten traumatischen Erlebnissen nicht zu untersuchen wären. Die gefundenen Ergebnisse dieser Dissertation erweitern bestehende Modelle zur Intrusionsentstehung durch neuroendokrinologische und physiologische Faktoren und tragen dadurch zu einem relevanten Wissenszuwachs bei. Die Ergebnisse geben Aufschluss über neuroendokrinologische und physiologische Aspekte unwillkürlicher intrusiver Erinnerungen und willkürlicher Traumaerinnerungen.

1 Theoretical and Empirical Background

Memories of traumatic life events belong to the category of "indestructible" memories as described by William James. Over a century later, the question raised by William James why some memories stay with us much longer than others cannot be answered satisfactorily. The biological mechanisms impacting on the formation of trauma related memories, such as intrusive memories and voluntary memory of trauma details, are still unclear.

Intrusive memories are a core feature of posttraumatic stress disorder (PTSD). PTSD has a prevalence rate of 1.1-5.7 % of the general population in Europe and the USA and affects more women than men (American Psychiatric Association, 2013; Kessler et al., 2005; Wittchen et al., 2011). PTSD impacts on endocrine (Pitman et al., 2012; Zoladz & Diamond, 2013) and physiological (Pitman et al., 2012; Pole, 2007) functions. To gain a better understanding of trauma related memories as well as PTSD and to further develop psychological and medical treatment options, it is of fundamental importance to assess neuropsychoendocrinological and psychophysiological influences. The aim of this dissertation project was to study the influence of the noradrenergic system (study I) and the hypothalamus–pituitary–adrenal (HPA) axis (study II) during a trauma film on intrusive memories. Further, the influence of the noradrenergic system and the HPA axis on memory of trauma film details (study III) was studied. Finally, the association of baseline heart rate variability (HRV) and intrusive memories (study IV) was studied. All four studies were conducted with a trauma film paradigm within an experimental approach.

In section 1.1 an introduction on stress and memory will be given. Next, in section 1.2 the body's neuroendocrine stress response will be outlined. Further, in section 1.3 intrusive memories will be defined and their underlying cognitive processes will be outlined. Aditionally, the current empirical background on the involvement of the stress systems in PTSD and the etiology of intrusive memories will be presented. Finally, in section 1.4 the physiological parameter HRV, an indicator of autonomic nervous system (ANS) activity (Chapleau & Sabharwal, 2011; Malik, 1996), and its relation to PTSD symptomatology will be described.

1.1 Stress and memory

During emotional experiences in life hormone and brain systems are activated and lead to the consolidation of new memories (Roozendaal & McGaugh, 2011). The release of glucocorticoids and noradrenaline during stress leads to emotional memory formation of stressful events (Schwabe & Wolf, 2013).

There is extensive evidence for the impact of the noradrenergic system on emotional memory consolidation (Roozendaal & McGaugh, 2011; Schwabe, Joëls, Roozendaal, Wolf, & Oitzl, 2012; van Stegeren, 2008). While endogenous sAA is positively associated with emotional memory (Segal & Cahill, 2009), pharmacological studies show that noradrenergic inhibition during encoding and consolidation results in impaired memory for emotional material and noradrenergic activation enhances memory for emotional material (Chamberlain & Robbins, 2013). For example, in healthy participants a single dose administration of clonidine impaired (Kuffel et al., 2014) and a single dose administration of yohimbine enhanced memory performance compared to placebo within a word list paradigm (Wingenfeld et al., 2013).

The HPA axis influences emotional memory formation during consolidation (Finsterwald & Alberini, 2014; Schwabe & Wolf, 2013). For example, participants who received hydrocortisone (synthetic cortisol) instead of placebo before encoding and consolidation of pictures showed enhanced memory for emotionally arousing pictures but not for neutral pictures 24 hours after consolidation (Kuhlmann & Wolf, 2006). Additionally, the interaction of enhanced activation of the HPA axis and the noradrenergic system during consolidation seems to be involved in emotional memory formation (Segal et al., 2014).

In sum, noradrenergic activation enhances emotional memory while noradrenergic inhibition inhibits emotional memory (Roozendaal & McGaugh, 2011). Further, cortisol enhances emotional memory consolidation (Schwabe & Wolf, 2013).

1.2 The neuroendocrine stress response

Exposure to a potential threat (stressor) of physical or psychological nature sets off a stress response with the aim of coping with the stressor and reinstating homeostasis, a dynamic and open equilibrium (de Kloet, Joëls, & Holsboer, 2005; Joels & Baram, 2009). The prefrontal cortex and limbic structures, particularly the hippocampus and the amygdala, are involved in the subjective appraisal of an event as stressful (Schwabe & Wolf, 2013). The hypothalamus is connected with the named brain regions and leads to a fast mobilization of the sympathetic nervous system (de Kloet, 2014; Sandi & Haller, 2015; Schwabe et al., 2012) and a slower mobilization of the HPA axis (Schwabe & Wolf, 2013).

The sympathetic nervous system orchestrates the organism's fight-or-flight response. Sympathetic preganglionic neurons are activated and project indirectly to end organs as well as to cells of the adrenal medulla. That results in a rapid release of the catecholamines adrenaline and noradrenaline (de Kloet et al., 2005; Ulrich-Lai & Herman, 2009). Noradrenaline[1] is primarily released from the locus coeruleus as well as brainstem sites and adrenaline is primarily released from the adrenal medullas (de Kloet et al., 2005; Schwabe & Wolf, 2013; Ulrich-Lai & Herman, 2009). Subsequently, heart rate increases and blood flow to the involved organs is enhanced (Schwabe et al., 2012).

The HPA axis initiates the release of corticotropin releasing factor (CRF) and vasopressin (AVP) into the portal vessel system. CRF, in turn, activates the pituitary and leads to the secretion of adrenocorticotropin (ACTH) (de Kloet et al., 2005; Lupien & Lepage, 2001). ACTH provokes an increased production and release of glucocorticoids, cortisol in humans and corticosterone in rodents, from the adrenal cortex into the blood circulation (de Kloet et al., 2005). Subsequently, the glucocorticoids enter the brain and bind to membrane-bound as well as

[1] Noradrenaline belongs to the group of catecholamines and acts as a hormone and neurotransmitter. Membrane potentials change rapidly as noradrenaline binds to alpha- and beta-adrenoceptors (Schwabe & Wolf, 2013).

[2] The mineralocorticoid receptor is involved in the appraisal phase and the beginning of the stress reaction (de Kloet et al., 2005).

intracellular mineralocorticoid receptors [2] and glucocorticoid receptors [3] (ter Heegde, de Rijk, & Vinkers, 2015). Through slow, genomic processes the glucocorticoids influence neuronal functions by affecting gene transcription (Datson, van der Perk, de Kloet, & Vreugdenhil, 2001). The affinity of glucocorticoids for mineralocorticoid receptors is ten times higher than for glucocorticoid receptors (Reul & de Kloet, 1985). The mineralocorticoid receptor is mainly expressed in limbic structures, such as in the hippocampus and the amygdala, whereas the glucocorticoid receptor is ubiquitously distributed throughout most brain regions, among others in the hippocampus, the paraventricular nucleus, the cerebral cortex, the limbic system, other hypothalamic and most brainstem monoaminergic nuclei (de Kloet et al., 2005; Lupien & Lepage, 2001; Reul & de Kloet, 1985). In addition, rapid, non-genomic processes on memory via membrane-associated glucocorticiod activity have been suggested by recent evidence (de Kloet, Karst, & Joels, 2008). The stress response of the HPA axis is regulated via negative feedback mechanisms depending on the concentration of circulating glucocorticoids in different brain areas, e.g. the hippocampus, the hypothalamus, and the pituitary (de Kloet et al., 2005). Cortisol blood levels peak 15-30 minutes after stress onset and decline to baseline levels within an hour (de Kloet, 2014). A simplified schematic representation of the body's noradrenaline and cortisol release is shown in Figure 1.

[2] The mineralocorticoid receptor is involved in the appraisal phase and the beginning of the stress reaction (de Kloet et al., 2005).

[3] The glucocorticoid receptor compared to the mineralocorticoid receptor is only activated after the release of larger glucocorticoid amounts and is implicated in the termination of the stress response, the mobilization of energy, and the recovery process. Further, the glucocorticoid receptor supports the storage of memory to prepare the organism for stressful events in the future (de Kloet et al., 2005).

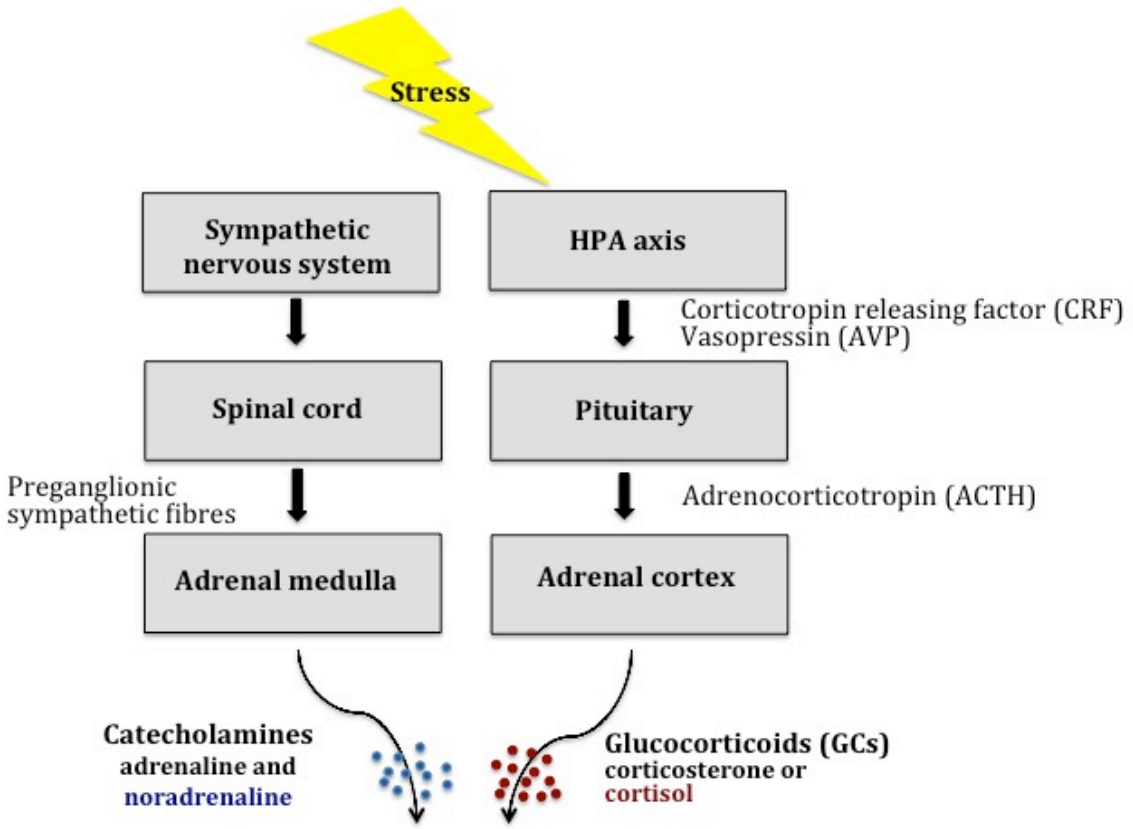

Figure 1. Simplified schematic representation of the stress response. On the left side the pathway of the sympathetic nervous system leading to the release of noradrenaline is depicted. On the right side the pathway of the HPA axis leading to the release of cortisol is depicted.

In conclusion, acute stress results in the mobilization of the HPA axis and the sympathetic nervous system (de Kloet, 2014; Sandi & Haller, 2015; Schwabe et al., 2012). Amongst other things, the release of cortisol and noradrenaline is increased as part of the endocrinological stress reaction.

1.3 Intrusive symptoms – a core feature of PTSD

During a lifetime the majority of the population is exposed to at least one traumatic event. However, only a minority of the exposed individuals develops PTSD (Kessler, Sonnega, Bromet, Hughes, & Nelson, 1995). A trauma is defined as "actual or threatened death, serious injury or sexual violence" (American Psychiatric Association, 2013). Inter alia, involuntary, intrusive, distressing, and recurrent

memories of that trauma are called intrusive symptoms and represent a core feature of PTSD (American Psychiatric Association, 2013). Intrusions shortly after trauma are of specific interest because they predict PTSD diagnosis later on (Ehlers, Hackmann, & Michael, 2004; Shalev, Peri, Canetti, & Schreiber, 1996). Intrusions are triggered, involuntary recollections related to the traumatic event and seem to appear spontaneously in consciousness (Brewin & Saunders, 2001; Holmes, James, Kilford, & Deeprose, 2010; Mace, 2008). While intrusions can appear as verbal thoughts and sensory based mental images, the latter represent the clinical phenomenology and are therefore of main interest (Holmes & Bourne, 2008). Further, intrusive and deliberately recalled emotional memories typically occur in form of sensory based mental images (Arntz, de Groot, & Kindt, 2005; Conway, 2001). Intrusive memories are experienced vividly, sometimes as if the trauma is happening again (Ehlers et al., 2004; Hackmann, Ehlers, Speckens, & Clark, 2004), and can include all physical sensations (Arntz et al., 2005; Conway, 2001), like the smell of gasoline after a traumatic car accident or the taste of sperm after sexual abuse. Further, an intrusive memory can take form of an emotion or physiological reexperience without recollection of the trauma (Schacter, Koutstaal, & Norman, 1997). However, intrusive memories are mostly visual (Ehlers & Steil, 1995; van der Kolk & Fisler, 1995), for example the victim of a car accident sees the bright lights of the approaching car. Intrusions can be triggered by temporally associated cues in the form of situations or stimuli (Ehlers & Clark, 2000).

It is widely accepted that intrusive symptoms are the result of trauma memory encoding, organization, and retrieval which deviate from normal autobiographical memory processing (Brewin, Dalgleish, & Joseph, 1996; Ehlers & Clark, 2000; Ehlers et al., 2004). The cognitive theory of PTSD by Ehlers and Clark (2000) and the dual representation theory by Brewin and colleagues (Brewin et al., 1996; Brewin, Gregory, Lipton, & Burgess, 2010) emphasize the importance of memory processes as well as encoding mechanisms for the development of intrusive memories and PTSD etiology (Holmes & Bourne, 2008). Cognitive behavioral therapy is based on these theories with similar statements regarding intrusion formation. In the cognitive theory of PTSD (Ehlers & Clark, 2000) it is assumed that memory of the trauma is not elaborated adequately nor integrated well in context terms, e.g. in time and place. Further, the theory postulates stronger

stimulus-stimulus associations and stimulus-response associations for traumatic memory. Finally, for trauma associated stimuli Ehlers and Clark (2000) propose stronger perceptual priming which leads to a reduced perceptual threshold of those stimuli.

The dual representation theory of PTSD (Brewin et al., 1996; Brewin et al., 2010) postulates the existence of two tightly associated separate memory mechanisms in healthy individuals. Sensory details as well as affections and emotions experienced during trauma are included in the sensory-bound memory representations (S-reps), which are activated automatically by specific cues in the enviroment. The other contextualized memory representations (C-reps) include an abstract description of the sensory input in addition to the experienced context (spatial and personal) of the individual and can be retrieved voluntarily or automatically. Very strong encoding of S-reps supports involuntary intrusive memories whereas C-reps support episodic memories and voluntary recall of traumatic memory.

1.3.1 The noradrenergic system, PTSD, and intrusive memories

Growing evidence from rodent and human studies on PTSD neurobiology shows an involvement of the noradrenergic system (Bailey, Cordell, Sobin, & Neumeister, 2013; Krystal & Neumeister, 2009). For example, after waking up salivary alpha-amylase (sAA) activity is increased in war refugees with PTSD diagnosis compared to control subjects, which indicates increased noradrenergic activation (van Stegeren, Rohleder, Everaerd, & Wolf, 2006). A possitive association of sAA secretion and reported PTSD symptoms was also found (Thoma, Joksimovic, Kirschbaum, Wolf, & Rohleder, 2012). Furthermore, 24 h urinary norepinephrine is increased in women with a history of sexual childhood abuse and PTSD diagnosis compared to nonabused women and women with a history of sexual childhood abuse but without PTSD diagnosis (Lemieux & Coe, 1995). Additionally, in women with PTSD less trauma-related nightmares were observed when noradrenergic activation was pharmacologically blocked with prazosin, an alpha-1

adrenergic antagonist[4], compared to female PTSD patients who received placebo (Taylor et al., 2006).

The noradrenergic system also seems to be associated with intrusive memories (Southwick et al., 1999). For example, a onetime administration of alpha-2 selective antagonist yohimbine, which enhances noradrenergic activation, or placebo on two separate test days in PTSD patients resulted in 40 percent of patients experiencing flashbacks on the day yohimbine was administered and five percent experiencing flashbacks on the day placebo was administered (Southwick et al., 1993). Additionally, the interaction of increased salivary cortisol levels and increased sAA activity during consolidation seems to be important as it predicts an increase of intrusions of emotionally negative images in PTSD patients (Nicholson, Bryant, & Felmingham, 2014) and healthy controls (Bryant, McGrath, & Felmingham, 2013). Further, within a trauma film paradigm salivary cortisol levels after watching a stressful film sequence were positively correlated with the subsequent number of intrusions in participants with increased sAA acitivity (Chou, La Marca, Steptoe, & Brewin, 2014). Sex differences in intrusion formation (Bryant et al., 2013; Felmingham, Tran, Fong, & Bryant, 2012) might occur due to sex-specific neurobiological stress mechanisms (Andreano & Cahill, 2009).

1.3.2 The HPA axis, PTSD, and intrusive memories

Patients with PTSD usually show decreased basal cortisol levels as a result of enhanced negative feedback sensitivity (Yehuda et al., 2015). Lower saliva cortisol in the mornings and higher saliva cortisol in the afternoons shortly after trauma were associated with subsequent PTSD (McFarlane, Barton, Yehuda, & Wittert, 2011). Further, female victims of sexual assault with lower serum cortisol levels 72 hours after trauma showed more PTSD symptoms six weeks later than women

[4] An antagonist is a substance that binds to a cellular receptor for a neurotransmitter, hormone, or another drug and thereby blocks its action without having a physiological effect itself (Neubig, Spedding, Kenakin, & Christopoulos, 2003).

with higher serum cortisol levels 72 hours after trauma (Walsh et al., 2013). Additionally, intensive care unit patients who received hydrocortisone treatment subsequently developed less PTSD symptoms (Schelling et al., 2006). At today's status of knowledge, hydrocortisone seems to be the only effective pharmacological intervention after trauma to prevent PTSD (Sijbrandij, Kleiboer, Bisson, Barbui, & Cuijpers, 2015).

Findings on cortisol and intrusions are contradictory. While one study found post accident trauma urinary cortisol to be associated with intrusive symptoms after six weeks in children (Delahanty, Nugent, Christopher, & Walsh, 2005), another study could not show an association of plasma, saliva, and urinary cortisol after trauma when intrusions were assessed five months later (Shalev et al., 2008). Initial treatments of PTSD patients with exogenous hydrocortisone resulted in an inhibition of trauma memories (Aerni et al., 2004). However, it was not possible to further confirm these findings as no effect of exogenous hydrocortisone was found on intrusions in women with PTSD (Ludäscher et al., 2015).

Taken together, intrusive memories occur due to different encoding, organization, and retrieval processes of trauma memory compared to non-traumatic autobiographical memory (Brewin et al., 1996; Ehlers & Clark, 2000; Ehlers et al., 2004). First evidence suggests an association of noradrenergic and HPA axis activation during consolidation and intrusive memories in healthy participants (Bryant et al., 2013; Chou et al., 2014).

1.4 Heart rate variability, trauma and PTSD

Heart rate variability represents the variation in the time intervals between consecutive heartbeats visualized in Figure 2 (Laborde, Mosley, & Thayer, 2017) and serves as an indicator of ANS activity (Chapleau & Sabharwal, 2011; Malik, 1996). Therefore, it has become a target of psychophysiological research (Laborde et al., 2017). The parasympathetic system with the vagus nerve as its main nerve and the sympathetic system are part of the ANS (Sammito & Bockelmann, 2015). When the body is at rest parasympathetic activity predominates, as reflected by

increased HRV, while the sympathetic system dominates during stimulating activities (Sammito & Bockelmann, 2015).

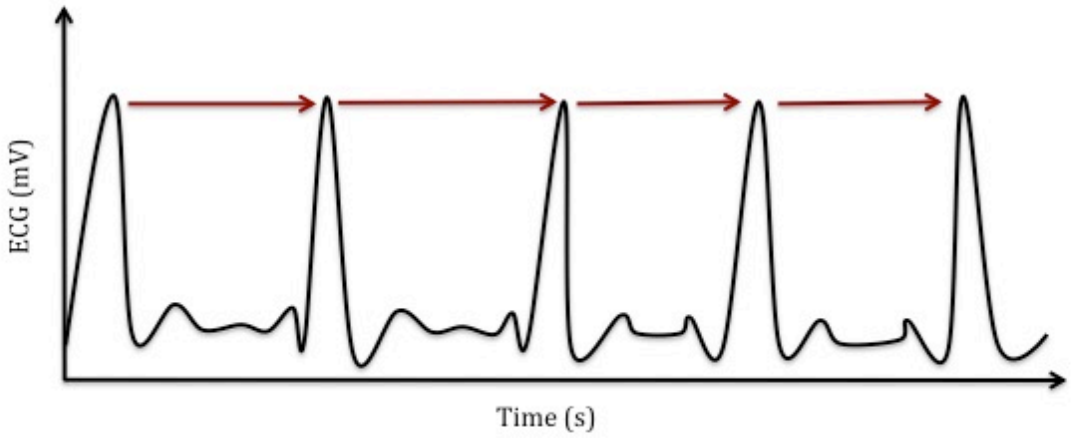

Figure 2. Consecutive heart beats. Heart rate variability represents the variation in the time intervals between consecutive heartbeats represented by the red arrows.

Various mathematically calculated parameters such as low-frequency (LF), high-frequency (HF), and low-frequency/high-frequency (LF/HF) ratio are combined under the term HRV and can be assigned to time-domain, frequency-domain, and non-linear methods of HRV-analysis (Bravi, Longtin, & Seely, 2011; Smith, Owen, & Reynolds, 2013a, b). LF/HF ratio is mostly interpreted as an indicator for sympathetic/parasympathetic balance (Heathers, 2014). However, this interpretation has been profoundly criticized (Billman, 2013) since LF power seems not only to reflect sympathetic but also parasympathetic activation (Malik, 1996). This leaves the physiological origin of LF/HF ratio unclear. Uncontroversial is the fact that HF power reflects parasympathetic activation (Malik, 1996).

Decreased baseline HRV in healthy individuals before military deployment is associated with increased PTSD symptoms after military deployment with trauma exposure (Minassian et al., 2015; Zucker, Samuelson, Muench, Greenberg, & Gevirtz, 2009). Cognitive control deficits, a risk factor for intrusion development, seem to be associated with reduced HRV in healthy individuals (Gillie, Vasey, & Thayer, 2014, 2015). Further, the Marine Resiliency Study found that reduced HRV before combat deployment in male marine soldiers was

associated with higher post-deployment PTSD rates (Minassian et al., 2015). This finding was replicated by another large prospective study, the Warriors Achieving Resilience Study, but only in Army National Guard soldiers with higher PTSD symptoms before deployment (Pyne et al., 2016).

Taken together, HRV serves as an indicator of ANS activity (Chapleau & Sabharwal, 2011; Malik, 1996). First evidence suggests that reduced baseline HRV is associated with increased post-trauma PTSD rates (Minassian et al., 2015; Pyne et al., 2016).

2 Aims and Design of the Dissertation Project

In this dissertation project the main objective is to examine underexplored neuropsychoendocrinological and autonomic influences on trauma film related memory to gain a better understanding of PTSD symptomatology, specifically the intrusions.

In study I (Rombold et al., 2016a) and II (Rombold et al., 2016b) the influences of the noradrenergic system and the HPA axis during encoding and consolidation of a trauma film on subsequent intrusion formation is assessed. In study III (Rombold-Bruehl et al., 2017a) noradrenergic and HPA axis influences during encoding and consolidation of a trauma film on subsequent forced choice recognition of trauma film details is examined. The sample of study III consists of all groups of study I and study II. In study IV (Rombold-Bruehl et al., 2017b) the association of baseline HRV and intrusive memories following the trauma film is examined. The sample of study IV consists of the combined placebo groups of study I and study II.

In section 2.1 the research questions and hypotheses for each study will be outlined. Subsequently, in section 2.2 the rational of the four studies will be explained in order to understand the design of the studies that address the research questions and test the hypotheses.

2.1 Research questions and hypotheses

In the following I will briefly outline the considerations and current state of research leading to the formulation of the research questions and the associated hypotheses for studies I, II, III, and IV.

2.1.1 Study I

As presented in sections 1.3 and 1.1.1, the noradrenergic system is involved in PTSD neurobiology and emotional memory formation. However, there is only scarce evidence of noradrenergic influences on intrusion formation and

experimental designs are lacking. To close this research gap, the potential influence of the noradrenergic system during traumatic stress (trauma film[5]) on the development of subsequent intrusive memories is assessed. The following research questions were raised.

Research questions

Does the inhibition or activation of the noradrenergic system during encoding and consolidation of a distressing event (trauma film) influence the *number* of consecutive intrusive memories of that event in healthy women?

Does the inhibition or activation of the noradrenergic system during encoding and consolidation of a distressing event (trauma film) influence the *vividness* of consecutive intrusive memories of that event in healthy women?

Does the inhibition or activation of the noradrenergic system during encoding and consolidation of a distressing event (trauma film) influence the *degree of distress* caused by the intrusions of that event in healthy women?

Hypotheses

Noradrenergic activation leads to an increased number of intrusive memories of the trauma film, more vivid intrusions, and more distressing intrusions of the trauma film compared with placebo and inhibited noradrenergic activity. Further, lower noradrenergic activity leads to a decreased number of intrusions, decreased vividness and decreased distress of intrusions compared with placebo and noradrenergic activation.

2.1.2 Study II

As presented in sections 1.1.2 and 1.3, several but not all studies indicate an association of post-trauma cortisol levels and PTSD symptoms. Although cortisol is involved in emotional memory formation, there is only scarce knowledge about the influence of cortisol on intrusive memory formation. Therefore, the potential

[5] In this dissertation the film sequence shown to the participant is called trauma film as part of a trauma film paradigm.

influence of cortisol during traumatic stress (trauma film) on the development of subsequent intrusive memories is assessed. To gain more knowledge about the influence of cortisol on intrusion formation, the following research questions were raised.

Research questions

Does cortisol during encoding and consolidation of a distressing event (trauma film) influence the *number* of consecutive intrusive memories in healthy women?

Does cortisol during encoding and consolidation of a distressing event (trauma film) influence the *vividness* of consecutive intrusive memories in healthy women?

Does cortisol during encoding and consolidation of a distressing event (trauma film) influence the *degree of distress* caused by the intrusions in healthy women?

Hypotheses

Hydrocortisone leads to an increased number of intrusive memories of the trauma film, more vivid intrusions, and more distressing intrusive memories of the trauma film compared with placebo.

2.1.3 Study III

As presented in section 1.3, the noradrenergic system and the HPA axis are involved in emotional memory formation. However, their influence during trauma on subsequent recognition of trauma details is unclear. Therefore, the potential influence of the noradrenergic system and the HPA axis during traumatic stress (trauma film) on subsequent forced choice recognition of trauma film details is assessed. The following research questions were raised.

Research questions

Does noradrenergic activity during a distressing event (trauma film) influence subsequent *memory for trauma film details* in healthy women?

Does HPA axis activation during a distressing event (trauma film) influence subsequent *memory for trauma film details* in healthy women?

Hypotheses

Inhibited noradrenergic activity impairs trauma film memory compared to placebo, exogenous cortisol and noradrenergic activation. Further, increased noradrenergic activity and exogenous cortisol enhance memory for the trauma film compared to placebo and inhibited noradrenergic activity. Finally, trauma film memory impairment and enhancement is specifically pronounced for peri-trauma film scene details, compared to pre-trauma film scene details and post-trauma film scene details.

2.1.4 Study IV

As presented in section 1.4, first evidence suggests an association of pre-trauma HRV and post-trauma PTSD symptomatology. Data on baseline HRV and post-stress intrusive memories within controlled laboratory conditions are lacking. Therefore, it is assessed whether HRV before traumatic stress (trauma film) predicts consecutive intrusive memories in healthy women within the placebo groups. To shed light on the impact of HRV on intrusion formation after trauma, the following research question was raised.

Research question

Does HRV at rest before a distressing event (trauma film) predict the *number* of consecutive intrusive memories in healthy women?

Hypothesis

Lower baseline HRV before the trauma film is associated with more consecutive intrusive memories of the trauma film.

2.2 Rationale of the studies

In this section the rational of the four studies will be outlined. The design of the studies, which intend to expand the current state of knowledge (section 1) and provide answers to the raised research questions (section 2.1), will be described. All studies were conducted at Charité – Universitätsmedizin Berlin, corporate member of Freie Universität Berlin, Humboldt-Universität zu Berlin, and Berlin Institute of Health, Campus Benjamin Franklin, and were approved by the ethics committee of the German Psychology Association (DGPs). All participants provided their written informed consent at least 24 hours before the assessment. Participants were asked to refrain from smoking, physical exercise, eating, consuming caffeine, and drinking alcohol or other beverages (except for water) at least one hour prior to the assessment. Each participant was tested on a separate day.

2.2.1 Trauma film paradigm

A trauma film paradigm is a suitable design to study psychological trauma and intrusive memories in the laboratory (Holmes & Bourne, 2008; James et al., 2016). The assessment day on which the trauma film was shown to the participants is visualized in Figure 3. Generally, film content of traumatic events is used to evoke symptoms which are analogous to the symptoms after real life trauma, like intrusive memories and physiological arousal (James et al., 2016). Experimentally-induced intrusive memories are shortlasting and disappear entirely within a week (Butler, Wells, & Dewick, 1995; Holmes, Brewin, & Hennessy, 2004; Weidmann, Conradi, Gröger, Fehm, & Fydrich, 2009). A sequence from the openly available film '*Irréversible*' (Gaspar Noé, 2002) has been shown to be especially suitable to induce intrusive memories in healthy participants (Weidmann et al., 2009). For all four studies of this thesis the same sequences from the film were cut out and put together so the participants could follow the sequences in the right chronological

order[6]. In the 14 minutes and 40 seconds long film sequence a women is having fun at a party with her partner and another male friend. After an argument with her partner she decides to leave the party by herself and rejects her friend's offer to accompany her home. Because of a busy road she has to take a pedestrian underpass where she gets threatened by a man with a knife, anally raped (for several screentime minutes), and brutally beaten until she is left on the ground unconsciously. In the last scene of the sequence her partner and her friend jokingly leave the party until they see the woman's bleeding body being carried into an ambulance car.

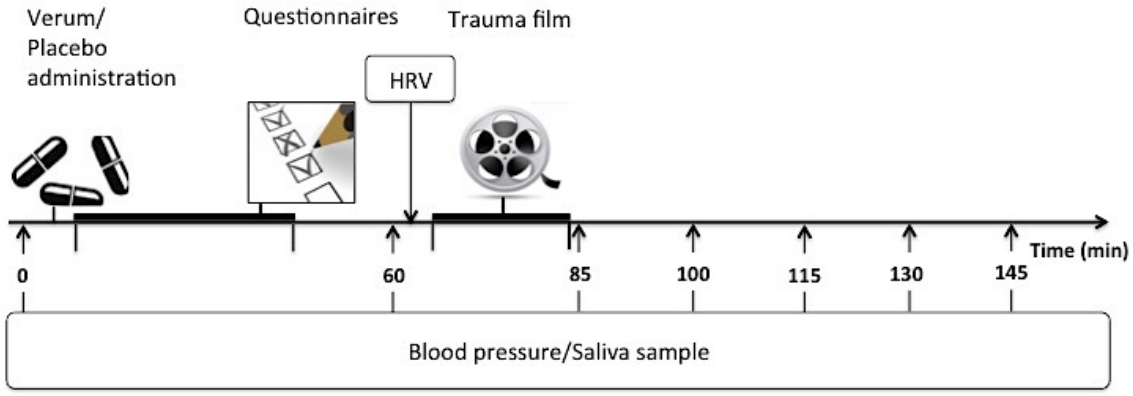

Figure 3. Procedure on the day of the trauma film. Abbreviation: HRV, heart rate variability.

2.2.2 Intrusion diary

Intrusive memories in studies I, II, and IV were recorded with a pencil and paper diary according to Holmes and colleagues (2004) and Weidmann and colleagues (2009). Additionally, an online diary with identical content to the pencil and paper diary was installed and participants had to transfer their notes from the pencil and paper version to the online version each night. This way participation was insured as the researcher could check the online diary inscriptions. A daily text message was sent to remind the participants to transfer the intrusions. Participants had to

[6] The film 'Irréversible' (Gaspar Noé, 2002) shows the plot backwards. For more methodical details on the presentation of the film please see method section of study I.

provide information about spontaneity of appearance, frequency, modality (image, thought based, or both), content, vividness, and the degree of distress. Vividness of the intrusions and degree of distress caused by the intrusions had to be rated between zero ('not at all') to five ('a lot'). Evaluation took place by summing up daily ratings. After the intrusion assessment participants returned the diary to the researcher who checked every intrusion for its intrusiveness by including only memories that occurred spontaneously, were image or image and thought based, and had vividness and degree of distress >1 (Arntz et al., 2005; Ehlers et al., 2004). This approach has a very good interrater reliability as shown by a study from Hagenaars and Arntz (2012) which used a similar intrusion assessment with an intraclass correlation of 0.997.

2.2.3 Participant recruitment

A total of 178 healthy, female university students were recruited via public postings, official university email lists, or social network university groups for this dissertation project (see section 3, 4, 5, and 6 for final sample sizes of each study). Pilot runs during preparation of this dissertation project with the trauma film also included male participants. However, it was observed that male participants reported different post film emotions like '*guilt*' and instead of identifying themselves with the female victim they rather identified themselves with a male witness of the rape who did not help the woman. Therefore, only women were recruited. The Structured Clinical Interview for DSM-IV Axis I Disorders (SCID-I) (First, Spitzer, Gibbon, & Williams, 1995; Wittchen, Fydrich, & Zaudig, 1997) was conducted over the phone by the SCID-I trained female researcher to exclude participants with former or present DSM-IV Axis I disorders. Further exclusion criteria were any physical illnesses or medication intake (except oral contraceptive), history of sexual abuse or rape, and lactation period or pregnancy. To control for the latter, the HCG ULTRA pregnancy test was implemented on the first test day. The women in all studies were between 18 and 44 years, spoke German on a native level, and received financial remuneration (35 Euro).

2.2.4 Yohimbine, Clonidine, and Hydrocortisone

Yohimbine is an indole alkaloid which is derived from the bark of a West African tree and is widely used for the treatment of sexual dysfunction in men. The alpha-2 adrenergic autoreceptors regulate the synaptic concentration of noradrenaline in the peripheral and central sympathetic system (Szabo, Hedler, & Starke, 1989). For study I and III yohimbine (10 mg) was chosen because it stimulates peripheral and central noradrenergic activity via inhibiting alpha-2 adrenergic autoreceptors (Charney, Woods, Goodman, & Heninger, 1987; Peskind et al., 1995). Peak plasma levels of oral yohimbine occur after 45–60 min and its elimination half-life is about 0.60 (S.D. = 0.25) h (Owen et al., 1987).

Clonidine is a chemical imidazoline compound (Regunathan, Meeley, & Reis, 1991) which is widely used for the treatment of hypertension (Giovannitti, Thoms, & Crawford, 2015). Clonidine is an alpha-2 adrenergic receptor agonist[7] and clonidine (0.15 mg, Clonistada ®) was chosen for studies I and III because it inhibits central noradrenergic activity (Giovannitti et al., 2015). Oral clonidine peaks after 60–90 min and its elimination half-life is six to 12 hours (Reid, 1981).

Hydrocortisone is a synthetic version of the endogenous glucocorticoid cortisol which is produced by the adrenal cortx. In study II hydrocortisone (10 mg, Galen ®) was administered to increase cortisol levels. Hydrocortisone peaks within one to two hours and its elimination half-life is 1.3 to 1.6 hours (Toothaker et al., 1982).

2.2.5 Randomization process

In study I and III participants were randomly allocated to the yohimbine, placebo, or clonidine group. In study II participants were randomly allocated to the hydrocortisone or placebo group. In study I and III the randomization of yohimbine, clonidine, and placebo was carried out by the internal pharmacy of the

[7] An agonist is a substance with affinity to a cellular receptor. Receptor activity is increased by conventional agonists and reduced by inverse agonists (Neubig et al., 2003).

Charité – Universitätsmedizin Berlin. In study II the randomization of hydrocortisone and placebo was created with the computer program '*research randomizer*' (Urbaniak & Plous, 2013). The randomization protocols were stored in a closed envelope which was opened at the end of the study.

2.2.6 Blinding

To ensure the double-blind design of studies, neither the participants nor the researchers knew if yohimbine, clonidine, or placebo in studies I and III or if hydrocortisone or placebo in study II had been administered. Identical looking tablets of hydrocortisone and placebo were stored in identical looking cups. The blinding process of yohimbine, clonidine, and placebo was more complicated and time-consuming since yohimbine and clonidine did not look identical in their original form and had to be put into capsules. Because the original administration form was changed from tablet to capsules the Charité pharmacy had to carry out tests to ensure the effect was not impacted.

2.2.7 Measurement of blood pressure

Dyastolic and systolic blood pressure were measured with the automatic device *boso medicus uno* from Bosch Sohn Germany. During measurement participants were seated in a standardized position and instructed not to move.

2.2.8 Measurement and analysis of heart rate variability

The heart rate monitor Polar RS800CX was used to measure HRV. Various studies have demonstrated that heart rate monitor recordings concur sufficiently with ECG system recordings (Quintana, Heathers, & Kemp, 2012; Wallen, Hasson, Theorell, Canlon, & Osika, 2012). As recommended by the manufacturer the textile electrode was held under running water and then the electrode belt was worn around the chest below the chest muscles. During recordings, participants rested comfortably without moving in a seated position. Both feet were on the floor and

both hands placed on the legs.

Artifacts of physiological or technical origin were detected with the Polar Precision Performance™ software. Filter power was set to "*moderate*" and minimum protection zone to six beats * min-1. Interpolation of degree zero was applied to replace the abnormal inter-beat intervals with a mean value computed from the previous and following normal inter-beat intervals. Kubios HRV version 2.0 software (Niskanen, Tarvainen, Ranta-Aho, & Karjalainen, 2004) was used for data analysis. From the frequency-domain analysis, LF (0.04-0.15 Hz) and HF (0.15-0.4 Hz) power were calculated using fast fourier transformation.

2.2.9 Salivary assessment

The following chapter has been published as complementary material in Rombold et al. (2016a) and was mainly written by Dr. Julian Hellmann-Regen.

After thawing, Salivettes® were centrifuged for two minutes at 1000 x g. Saliva samples were always kept on ice and immediately subjected to both cortisol and amylase activity assays. Free cortisol was analyzed using a commercially available TR-FRET-based, in-house adopted immunoassay (Cisbio International, Codolet, France), which was performed in principle according to the manufacturer's instructions. In brief, two parts of sample were subjected to a fluorescence microtiter plate and one part of D2-conjugated cortisol was added immediately thereafter. Both components (saliva and D2-conjugate) were thoroughly mixed using a multi-channel pipette and centrifuged for two minutes at 1000 x g using a microtiter plate centrifuge (Heraeus Biofuge, Thermo Fisher Scientific, Braunschweig, Germany). After centrifugation, one part of Europium-cryptate-labelled anti-cortisol antibody was added, again thoroughly mixed, centrifuged (2 minutes at 1000 x g), and allowed to incubate for at least two hours. Appropriate authentic standards, negative, positive, and blank controls were included according to the manufacturer's instructions. After incubation, time-resolved fluorescence was measured at 620 nm and 665 nm using a Clariostar multimode plate reader (BMG Labtech, Ortenberg, Germany). Increase in fluorescence at 665 nm (acceptor fluorescence) was normalized to fluorescence at 620 nm (donor

fluorescence) to account for differences in plating volumes or micro bubbles, and calculated as relative increase in fluorescence over cryptate-only containing blanks. Inter- and intraassay coefficients of variation were below 12 %. All samples and standards were measured in duplicates.

Alpha-amylase activity was determined using a modified protocol of a previously published direct alpha-amylase assay (Lorentz, Gütschow, & Renner, 1999). The assay principle follows an IFCC method using 2-chloro-4-nitro-phenyl-a-D-maltotrioside (CNPG) as the chromogenic substrate and was adapted to be run at room temperature using six authentic alpha-amylase standards (Sigma-Aldrich, Taufkirchen, Germany) for absolute quantification. In brief, samples and standards were diluted 1:200 and one part of sample/standard was subjected to a clear microtiter plate. Plates were subsequently allowed to equilibrate to 20 °C inside the temperature-controlled multimode plate-reader and 20 parts of CNPG-containing chromogenic substrate (substrate start procedure) were injected in a time-controlled manner. Following each automated injection, the plate was shaken and an increase in absorbance was read at 405 nm periodically after exactly two, four, six, and eight minutes in each well. A linear increase in absorbance was assured for all samples and average increase in absorbance per minute (delta OD405 / min) was calculated for all samples and standards. Absolute quantification of alpha-amylase activity in samples was realized by four-parameter nonlinear regression analysis of the calibration standards ($r2 > 0,998$). Inter- and intraassay coefficients of variation were both lower than ten percent for alpha-amylase activity. All samples and standards were measured in triplicates.

2.2.10 Selection of psychological tests

The Childhood Trauma Questionnaire (CTQ) (Bernstein & Fink, 1998; Wingenfeld et al., 2010) was applied in studies I, II, and III to control for different childhood events as childhood trauma has been shown to be associated with noradrenergic stress responses (Otte et al., 2005) and an increased PTSD risk after trauma in adulthood (Breslau, Chilcoat, Kessler, & Davis, 1999). The trait scale of the State-Trait Anxiety Inventory (STAI-T) (Laux, Glanzmann, Schaffner, & Spielberger, 1981; Spielberger & Gorsuch, 1983) was utilized in studies I , II, and IV because of

a positive association of PTSD and pre-trauma trait anxiety (McNally et al., 2011) and since trait anxiety is associated with dysfunctions in the ANS (Rajcani, Solarikova, & Brezina, 2017). The Beck Depression Inventory (BDI) (Beck, 1978; Hautzinger, Bailer, Worall, & Keller, 1995) was utilized in study IV to assess depressive symptoms since reduced vagal activity has been found in depressed patients (Kemp et al., 2010). The Dissociation-Tension-Scale acute (DSS-Acute) (Stiglmayr, Braakmann, Haaf, Stieglitz, & Bohus, 2003) was applied in studies III and IV since dissociation during traumatic events is associated with disorganisation and disintegration of trauma details recall (Bedard-Gilligan & Zoellner, 2012) and increased ANS functioning (Kuhn, Blanchard, Fuse, Hickling, & Broderick, 2006; Ladwig et al., 2002; Nixon, Bryant, Moulds, Felmingham, & Mastrodomenico, 2005). The Impact of Event Questionnaire (IES) (Horowitz, Wilner, & Alvarez, 1979; Maercker & Schützwohl, 1998) was applied after seven days in studies I and II to evaluate PTSD-related symptoms, namely intrusion, avoidance, and hyperarousal. A 24-item forced choice recognition test was applied which was developed for study III to assess memory of the film. Four right and four wrong statements on film details before the physical and sexual violence, during the physical and sexual violence, and after the physical and sexual violence had to be answered. Participants had to decide if each of the 24 statements about the distressing film was right or wrong.

After the rationale of this dissertation has been outlined, the four studies will now be presented in the following.

3 Influence of the Noradrenergic System on the Formation of Intrusive Memories: An Experimental Approach with a Trauma Film Paradigm (study I)

This chapter has been published as 'Rombold, F., Wingenfeld, K., Renneberg, B., Hellmann-Regen, J., Otte, C., & Roepke, S. (2015). Influence of the noradrenergic system on the formation of intrusive memories in women: an experimental approach with a trauma film paradigm. *Psychological Medicine*. 46(12), 2523-2534'.

DOI: https://doi.org/10.1017/S0033291716001379

3.1 Abstract

Introduction: Intrusive memories of traumatic events are a core feature of PTSD but little is known about the neurobiological formation of intrusions. The aim of this study was to determine whether the activity of the noradrenergic system during an intrusion-inducing stressor would influence subsequent intrusive memories.

Methods: We conducted an experimental, double-blind, placebo-controlled study in 118 healthy women. Participants received a single dose of either 10 mg yohimbine, stimulating noradrenergic activity, or 0.15 mg clonidine, inhibiting noradrenergic activity, or placebo. Subsequently, they watched an established trauma film that induced short lasting intrusions. The number of consecutive intrusions resulting from the trauma film, the vividness of the intrusions, and the degree of distress evoked by the intrusions were assessed during the following four days. Salivary cortisol and alpha-amylase were collected at seven time points before and after the trauma film.

Results: A significant time by treatment interaction for the number of intrusions and the vividness of intrusions indicated a different time course of intrusions depending on treatment. Post-hoc tests revealed a delayed decrease of intrusive memories and a delayed decrease of intrusion vividness after the trauma film in the yohimbine group compared to the clonidine and placebo group. Furthermore, after yohimbine administration, a significant increase in salivary cortisol levels was observed during the trauma film.

Conclusions: Our findings indicate that pharmacological activation of the noradrenergic system during an emotionally negative event impacts on consecutive intrusive memories and their vividness. The noradrenergic system seems to be involved in the formation of intrusive memories.

3.2 Introduction

Posttraumatic stress disorder can develop after a traumatic stressor and it affects approximately 5.7 percent of the general population in the US (Kessler et al., 2005) and 1.1–2.9 percent in Europe (Wittchen et al., 2011). Intrusive symptoms are a core feature of PTSD comprising, inter alia, recurrent, involuntary, and intrusive distressing traumatic memories (American Psychiatric Association, 2013).

Rodent and human studies provide cumulative evidence for the involvement of the noradrenergic system in PTSD neurobiology (Bailey et al., 2013; Krystal & Neumeister, 2009). A recently published study found that medical outpatients from two Veterans Affairs medical centers with PTSD diagnosis show increased 24 hour urinary norepinephrine compared to medical outpatients without PTSD diagnosis (Wingenfeld, Whooley, Neylan, Otte, & Cohen, 2015). Furthermore, war refugee PTSD patients show increased sAA activity after awakening compared to healthy controls, and sAA secretion is positively associated with reported PTSD symptoms (Thoma et al., 2012). In addition, pharmacological interventions with the alpha-1 adrenergic antagonist prazosin, which blocks noradrenergic activation, have been shown to reduce distressing dreams, sleep disturbance, re-experiencing, avoidance, and hyperarousal compared to placebo in patients with PTSD (Raskind et al., 2003).

However, little is known about the neurobiological influences on the formation of intrusions after traumatic stress. The noradrenergic system could be involved, as indicated by studies in which a relationship between increased norepinephrine and intrusive memories has been found (Southwick et al., 1999). For example, in a study conducted by Southwick et al (1993), PTSD patients were administered a single dose of yohimbine, an alpha-2 selective antagonist that enhances noradrenergic activation, and placebo, each on a separate test day in a double blind, randomized balanced order. 40 percent of patients reported flashbacks in the yohimbine condition, while only five percent reported flashbacks in the placebo condition during the test day. Furthermore, in healthy men, it has been found that the interaction of increased sAA activity and increased salivary cortisol levels during the consolidation of emotionally negative images predicts an increase in intrusive memories (Bryant et al., 2013). This result has also been

found in PTSD patients compared to trauma exposed participants without PTSD and healthy controls (Nicholson et al., 2014).

The lack of suitable experimental designs to examine the neurobiology of the formation of intrusive memories has so far limited the number of studies. Recently, trauma film paradigms have been shown to induce short lasting intrusive memories in healthy individuals (Holmes & Bourne, 2008). The only study that has, so far, examined associations among salivary cortisol, sAA, and intrusion formation within a trauma film paradigm found a positive correlation between post-film salivary cortisol levels and the frequency of intrusions, however, only in individuals with increased sAA activity (Chou et al., 2014).

In contrast to the scarce evidence regarding the influence of the noradrenergic system on intrusion formation, there is a large number of studies investigating the influence of the noradrenergic system on other aspects of memory and emotional learning in humans (Roozendaal & McGaugh, 2011; Schwabe et al., 2012; van Stegeren, 2008). The administration of adrenergic receptor blockers (e.g., propranolol) leads to impaired memory performance for emotional events and stimuli in humans, while pharmacological noradrenergic activation (e.g., yohimbine) during encoding and consolidation enhances memory for emotional stimuli in the majority of studies (Chamberlain & Robbins, 2013). A positive association between sAA, assessed shortly after the presentation of emotionally arousing pictures, and memory of these pictures one week later has been found in healthy participants (Segal & Cahill, 2009).

In addition to the catecholaminergic system, the HPA axis releases cortisol when it is activated, and impacts the formation of emotional memory during encoding and consolidation in healthy individuals (Finsterwald & Alberini, 2014; Schwabe et al., 2012). For instance, elevated cortisol levels during encoding and consolidation after administration of hydrocortisone improve memory performance for emotionally arousing pictures compared to neutral pictures (Kuhlmann & Wolf, 2006). Therefore, cortisol levels during trauma might be relevant in formation of intrusions.

The aim of the current study was to examine the potential influence of noradrenergic activity during traumatic stress (trauma film) in healthy women on the consecutive development of intrusive memories in an experimental design. We

hypothesized that noradrenergic activation would lead to an increased number of intrusive memories of the trauma film, more vivid intrusions, and more distressing intrusions of the trauma film compared to placebo and inhibited noradrenergic activity. In addition, it was hypothesized that lower noradrenergic activity would lead to a decreased number of intrusions, decreased vividness and decreased distress of intrusions compared to placebo.

3.3 Methods

3.3.1 Participants

118 healthy university students were recruited via official university email lists or public postings. As the rape victim in the trauma film is female, the sample was restricted to women to increase homogeneity of our sample. Furthermore, pilot runs with the trauma film including male and female participants had shown that men did not identify themselves with the victim but rather with a male witness. Exclusion criteria included former or present DSM-IV Axis I disorders assessed by the SCID-I (First et al., 1995), physical illnesses, any medication intake (except oral contraceptive), history of sexual abuse or rape, and pregnancy or lactation period. The HCG ULTRA pregnancy test was implemented to exclude pregnancy. All women were between 18 and 44 years old (see Table 1) and spoke German on a native level. Out of the 118 participants, four women were excluded: two participants withdrew from the study after having watched the film for several minutes, one participant was excluded due to fatigue during the session after taking clonidine, and one participant was excluded due to missing diary data. This resulted in the final sample size of N = 114. All participants received financial remuneration (35 Euro).

Table 1. *Sample characteristics*

Characteristic	Clonidine *n* = 38 **M(SD) or n(%)**	Placebo *n* = 38 **M(SD) or n(%)**	Yohimbine *n* = 38 **M(SD) or n(%)**	Statistics	*p*
Age	23.3(3.6)	23.1(3.2)	23.4(4.5)	$F_{df2,111}$ = .08	.93
Intake of oral contraceptives	19(50.0 %)	22(57.9 %)	13(34.2 %)	χ^2 (2) = 4.43	.11
Current smoker	8(21.1 %)	4(10.5 %)	9(23.7 %)	χ^2 (2) = 2.45	.29
Menstrual cycle,				χ^2 (2) = 1.02	.62
follicular vs.	18(50 %)	18(47.4 %)	20(58.8 %)		
luteal phase	18(50 %)	20(52.6 %)	14(41.2 %)		
BMI	21.2(2.6)	22.4(3.2)	21.8(3.0)	$F_{df2,110}$ = 1.57	.21
CTQ	37.4(9.1)	38.3(10.0)	36.1(6.1)	$F_{df2,111}$ = .66	.52
STAI-T	37.2(7.0)	37.6(7.1)	35.7(6.8)	$F_{df2,111}$ = .81	.45
Participants who have seen the film before	2(1.8 %)	4(3.5 %)	1(0.9 %)	χ^2 (2) = 2.24	.33

Abbreviations: BMI, Body Mass Index; CTQ, Childhood Trauma Questionnaire; STAI-T, trait scale of the State-Trait Anxiety Inventory.

3.3.2 Procedure

The randomized, double-blind, placebo-controlled study was conducted at the Department of Psychiatry and Psychotherapy, Campus Benjamin Franklin, Charité – Universitätsmedizin Berlin, and was approved by the local ethics committee of the German Psychology Association (DGPs). Participants were asked to refrain from smoking, physical exercise, eating, consuming caffeine, and drinking alcohol or other beverages (except for water) at least one hour prior to the assessment. Written informed consent was obtained at least 24 hours before the assessment. Each participant was tested on a separate day.

Experimental phase

Participants were randomly assigned to either clonidine (0.15 mg), inhibiting noradrenergic activity, placebo, or yohimbine (10 mg), stimulating noradrenergic activity. The pills for both drugs as well as the placebo looked identical, hence ensuring that the experimenter was also blind to the experimental condition. Administration was conducted sixty minutes before the trauma film, since peak plasma levels of clonidine occur after 60-90 minutes (Reid, 1981) and yohimbine peaks in plasma after 45-60 minutes (Owen et al., 1987). Potential effects of the medication and the trauma film on salivary cortisol levels, sAA activity and blood pressure were measured at seven time points during the study. To do so, saliva was collected and blood pressure was measured by an automatic device (boso medicus uno, Bosch + Sohn Germany) at baseline, after medication intake, and five times after the trauma film. The CTQ (Bernstein & Fink, 1998; Wingenfeld et al., 2010) was utilized to control for differences in experienced childhood events, as childhood trauma has been shown to be associated to noradrenergic responses to stress (Otte et al., 2005) and to an increased risk of PTSD after trauma in adulthood (Breslau et al., 1999). Since PTSD is positively related to pre-trauma trait anxiety (McNally et al., 2011), the STAI-T (Laux et al., 1981; Spielberger & Gorsuch, 1983) was applied. After the assessment, participants completed the intrusion diary for the next four days. Participants were instructed to not talk to other potential participants about the content of the study.

Follow-up session

After seven days, the diary was returned and the IES (Horowitz et al., 1979; Maercker & Schützwohl, 1998) was applied to evaluate PTSD related symptoms: intrusion, avoidance and hyperarousal. In order to not inform participants about the trauma film before the experimental phase, they were asked at the end of the study if they had seen the film '*Irréversible*' before. After four weeks the participants were contacted and the IES was conducted again via phone. At the end of the study a debriefing was provided via phone and in written form.

3.3.3 Trauma film and intrusion diary

The trauma film paradigm was adopted, using a film scene which is particularly suitable for evoking short lasting intrusions in healthy participants and which has been used in previous studies (Weidmann et al., 2009). The scene from the film '*Irréversible*' (Gaspar Noé, 2002) was shown to all participants, depicting a scene (14 minutes, 40 seconds) in which a woman is raped by a man. The film was shown on a 2 x 2.5 meter screen. The sound was played through headphones. The female researcher was present while the participant watched the film.

A pencil and paper diary (Holmes et al., 2004; Weidmann et al., 2009) was used to record intrusions throughout the following four days. Participants were instructed to "record any spontaneously occurring memory from the trauma film" instantaneously after occurrence. The difference between image and thought based memories was explained to the participant. According to Holmes and colleagues (2004) memories in the diary were specified by spontaneity of appearance, frequency, modality (image, thought based, or both), content, vividness, and the degree of distress. Vividness and degree of distress of every single intrusion were rated from zero ("not at all") to five ("a lot") and evaluated by summing up daily ratings. A text message was sent at nine p.m. daily to remind participants to transfer inscriptions from their pencil and paper diary to an identical online diary to assure participation. When participants turned in the diary, the researcher went through every single memory with them and rated whether the respective memory was intrusive or not. Only memories which occurred spontaneously and included imagery with vividness and degree of distress > zero were considered intrusive and included in the analyses (Arntz et al., 2005; Conway, 2001; Ehlers et al., 2004; Hackmann et al., 2004).

3.3.4 Salivary assessment

Saliva samples were collected in order to determine salivary free cortisol and sAA (Salivettes®, blue cap; Sarstedt). To ensure homogenous saliva collection from all salivary glands, participants were instructed to move the cotton swab in their mouth in a circular pattern for one minute without actively chewing on it (Nater &

Rohleder, 2009). Samples were subsequently stored at -20 °C before biochemical analyses were performed in the Neurobiology Laboratory of the Department of Psychiatry, Charité, Universitätsmedizin Berlin. Salivary cortisol was analyzed using a commercially available TR-FRET-based, in-house adopted immunoassay (Cisbio International, Codolet, France), which was performed in principle according to the manufacturer's instructions (see section 2.2.8 for details). Inter- and intra-assay coefficients of variation were below 12 %. SAA activity was determined using a modified protocol of a previously published direct alpha-amylase assay (Lorentz et al., 1999). Inter- and intra-assay coefficients of variation were both lower than ten percent for sAA activity.

3.3.5 Statistical analysis

Statistical analyses were performed using SPSS Version 22.0. Statistical significance refers to a p value < 0.05. Differences between treatment groups in demographic baseline variables and in the conducted questionnaires during the study were tested by univariate analysis of variance (ANOVA) for continuous data and Pearson's chi-squared test for categorical data.

Three sets of repeated measures mixed design ANOVAs were used to test the effects of treatment (between-subjects factor) and time (within-subjects variable with seven levels) on salivary cortisol levels, sAA, and blood pressure. Furthermore, ANOVA was conducted to examine the effects of treatment (between-subjects factor) and time (within-subjects variable with four levels) on number of intrusions, vividness, and degree of distress as dependent variables. In all ANOVAs, homogeneity of variance was assessed by Levene's statistic and sphericity was examined with Mauchly's test. Since the assumption of sphericity was never met, Greenhouse–Geisser corrected p values are reported. For post-hoc comparisons, a Bonferroni correction for multiple comparisons was performed.

3.4 Results

3.4.1 Participant characteristics

The three groups did not differ in any of the demographic variables or possible confounders (see Table 1).

3.4.2 Salivary alpha-amylase and blood pressure

Salivary alpha-amylase activity throughout the assessment is displayed in Figure 4. There was a significant effect of time ($F_{df3.98,\ 421.71} = 13.57$, $p < .01$) as well as a significant effect of treatment ($F_{df2,106} = 7.41$, $p < .01$). Furthermore, a significant time x treatment interaction ($F_{df7.96,421.71} = 3.14$, $p < .01$) was revealed. Groups did not differ at baseline ($p > .05$). Post-hoc tests revealed that yohimbine led to higher sAA activity compared to placebo and clonidine ($p < .05$).

There was a significant effect of time and treatment for diastolic ($F_{df3.91,430.57} = 10.65$, $p < .01$, $F_{df2,110} = 33.36$, $p < .01$) and systolic ($F_{df4.13,454.52} = 16.43$, $p < .01$, $F_{df2,110} = 28.42$, $p < .01$) blood pressure. The interaction effect time x treatment was significant for diastolic ($F_{df7.83,430.57} = 11.16$, $p < .01$) and systolic blood pressure ($F_{df8.26,454.52} = 8.35$, $p < .01$), indicating that clonidine decreased diastolic ($p < .01$) and systolic ($p < .01$) blood pressure compared to yohimbine and placebo.

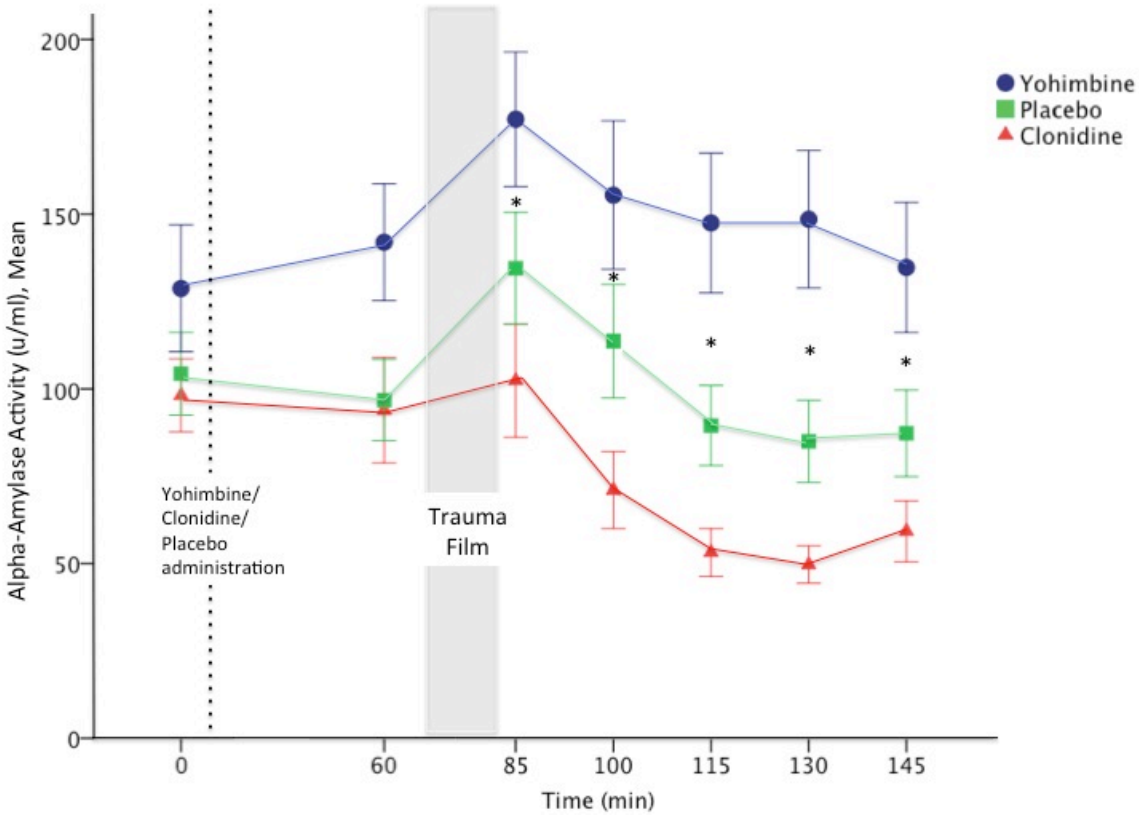

Figure 4. Salivary alpha-amylase activity at baseline and before and after the trauma film for the clonidine, placebo, and yohimbine group. Significant differences between the yohimbine and placebo group, and the yohimbine and clonidine group (* $p < .05$). Error bars represent +/- 1 SE.

3.4.3 Salivary cortisol

Salivary cortisol levels for the three experimental groups are displayed in Figure 5. While there was no treatment effect ($F_{df2,107} = 2.00$, $p = .15$), the effect of time was significant ($F_{df2.00,214.29} = 3.52$, $p < .01$). Furthermore, the time x treatment interaction was significant ($F_{df4.01,\ 214.29} = 3.37$, $p = .01$). Post-hoc tests revealed no difference in salivary cortisol levels between the three groups at baseline ($p > .05$). Furthermore, post-hoc tests revealed higher salivary cortisol levels in the yohimbine group compared to the placebo group ($p < .05$) and also compared to the clonidine group on a trend level ($p < .1$).

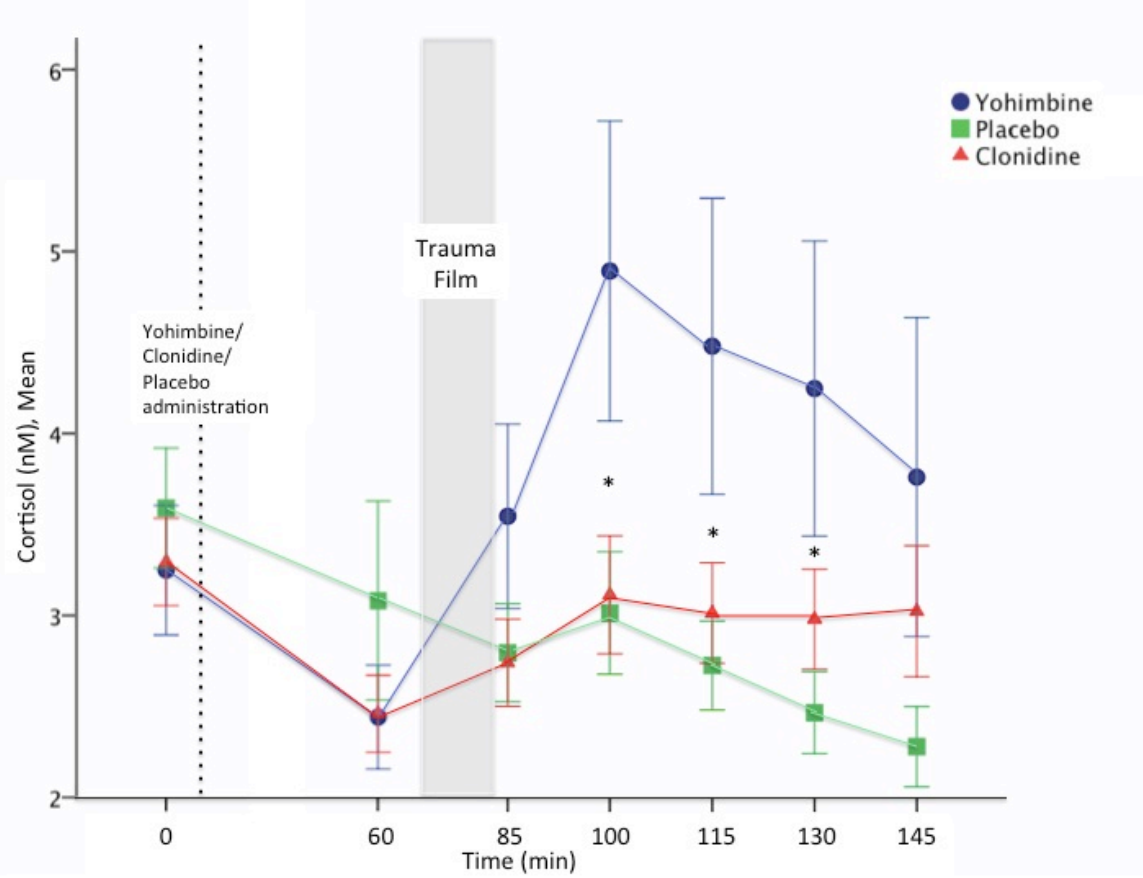

Figure 5. Salivary Cortisol levels at baseline and before and after the trauma film for the clonidine, placebo, and yohimbine group (* $p < .05$). Error bars represent +/- 1 SE.

3.4.4 Intrusions

As expected, the number of intrusions declined over time ($F_{df2.47,273.81} = 65.24$, $p < .01$). While there was no significant effect of treatment ($F_{df2,111} = 1.00$, $p = .37$), a significant time x treatment interaction was observed ($F_{df4.93,3.0} = 2.97$, $p = .01$), indicating a delayed decrease in the number of intrusive memories in the yohimbine group compared to the clonidine and placebo group. The interaction is displayed in Figure 6. Post-hoc tests revealed that participants in the yohimbine group reported more intrusive memories of the trauma film on the first day after the trauma film than participants in the clonidine ($p = .02$) and placebo ($p = .03$) group.

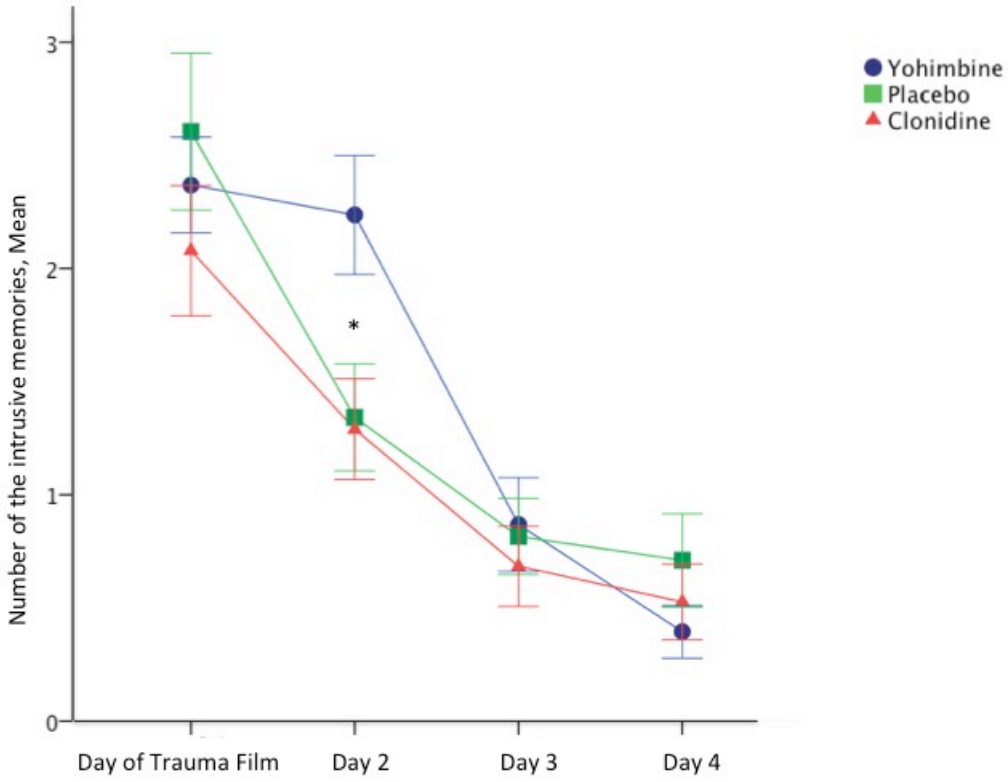

Figure 6. Results for the number of the intrusive memories for the yohimbine, placebo, and clonidine group over four days, starting on the day when the trauma film was shown to the participants (* $p < .05$). Error bars represent +/- 1 SE.

As expected, the vividness of intrusive memories also declined over time ($F_{df2.38,264.55} = 32.32$, $p < .01$). While there was no significant effect of treatment ($F_{df2,111} = 1.34$, $p = .27$), the time x treatment interaction was significant ($F_{df4.77,264.55} = 2.55$, $p = .03$), indicating a delayed decrease in the vividness of intrusive memories in the yohimbine group compared to the clonidine and placebo group. The interaction is displayed in Figure 7. Again, on the first day after the trauma film the reported vividness of the intrusions in the yohimbine group was significantly higher than in the placebo ($p = .03$) and clonidine ($p = .02$) group.

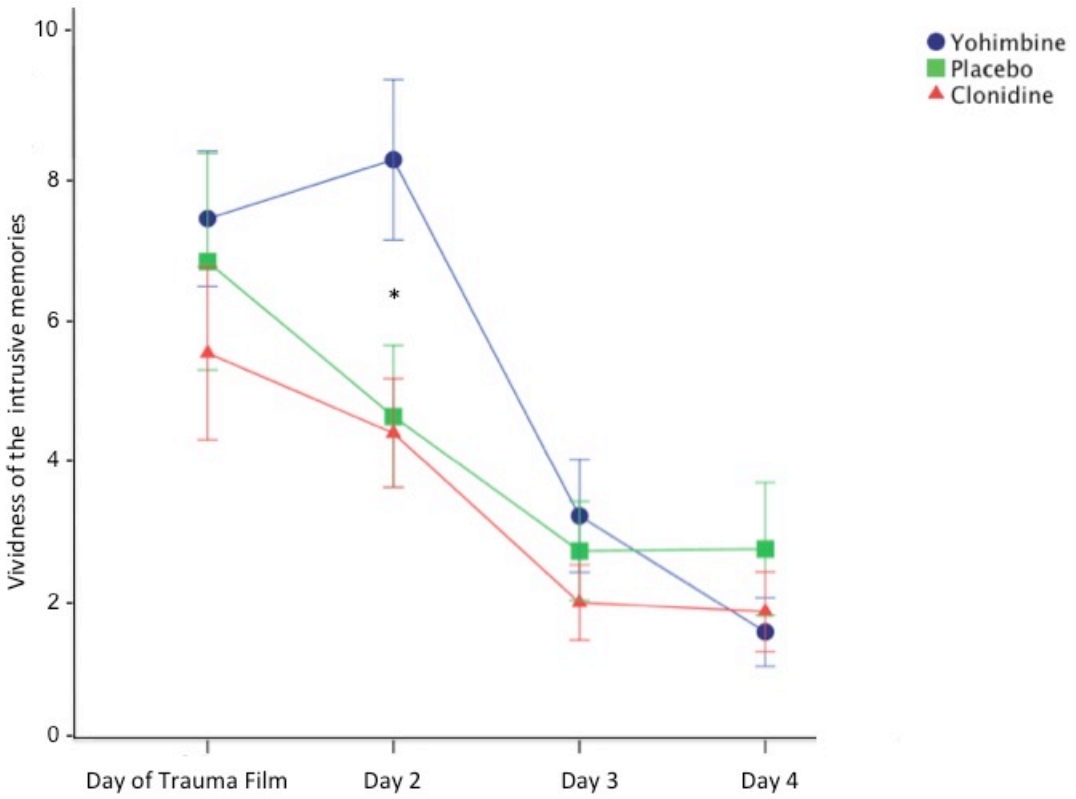

Figure 7. Results for the vividness of the intrusive memories for the yohimbine, placebo, and clonidine group over four days, starting on the day when the trauma film was shown to the participants (* $p < .05$). Error bars represent +/- 1 SE.

Yohimbine, clonidine, and placebo did not affect the degree of distress participants experienced through the intrusive memories (no effect of treatment, $F_{df2,111} = 1.17$, $p = .31$ and no treatment x time interaction, $F_{df4.76,264.40} = 1.73$, $p = .11$). However, there was a significant effect of time ($F_{df2.39,264.40} = 59.84$, $p < .01$) indicating a decline of distress over the course of four days.

Including sAA activity and cortisol levels (area under curves), childhood trauma (Bernstein & Fink, 1998; Wingenfeld et al., 2010), or intake of oral contraceptives separately as covariates in the general linear model did not modify the significance of the interaction effect (time x treatment) for the number of intrusions and vividness.

3.4.5 The Impact of Event Questionnaire

The IES (Horowitz et al., 1979; Maercker & Schützwohl, 1998) did not show any differences in PTSD related symptoms between the yohimbine, clonidine, and placebo group one week ($F_{df2,108} = .25$, $p = .78$) and four weeks after the assessment ($F_{df2,107} = .32$, $p = .72$).

3.5 Conclusions

We examined the influence of the noradrenergic system on encoding and consolidation of potentially intrusive memories after a trauma film paradigm in healthy women. Prior to the trauma film, participants received either yohimbine to stimulate noradrenergic activity, or clonidine to inhibit noradrenergic activity, or placebo. Intrusions occurring throughout the following four days were recorded.
After yohimbine the number of intrusive memories as well as their vividness showed a delayed decrease compared to clonidine and placebo over the four days following the trauma film. Our findings suggest that noradrenergic activation during encoding and consolidation of a traumatic event delays the decline of the frequency and vividness of intrusive memories. The current results may implicate that trauma survivors with higher noradrenergic activity during the traumatic event and/or during following consolidation are more likely to subsequently develop PTSD symptoms, i.e., intrusions. These findings are relevant because it has been shown that intrusions during the days and weeks after a trauma predict a PTSD diagnosis later on (Ehlers, 2010; Shalev et al., 1996).

Our results extend recent findings of a study by Bryant et al (2013), which showed that healthy participants reported more unintentional memories of depicted negative images after being stressed during consolidation compared to non-stressed participants. In this study, an interaction of increased salivary cortisol levels and sAA activity during consolidation predicted more unintentional memories in men but not in women (Bryant et al., 2013). So far, a single study has examined the influence of sAA and salivary cortisol during encoding and consolidation on subsequent intrusions within a trauma film paradigm but without manipulating glucocorticoid and noradrenaline signaling (Chou et al., 2014).

However, in this study, sAA activity during encoding and consolidation did not predict the vividness of subsequent intrusions in men and women. Also, peri-film sAA activity was not associated with the number of intrusions. However, a positive correlation between post-film salivary cortisol levels and the number of intrusions was found in individuals with increased sAA activity (Chou et al., 2014).

A strong increase in salivary cortisol levels 15 minutes after the end of the trauma film in the yohimbine condition was found. Contradictory findings exist on the influence of yohimbine administration on salivary cortisol levels in healthy controls. Some findings indicate enhanced salivary cortisol levels after yohimbine administration (Sommer et al., 2011) and others indicate no effect of yohimbine on salivary cortisol levels in controls (Gurguis, Vitton, & Uhde, 1997). In our study, noradrenergic activation after yohimbine during the trauma film might have served as a facilitator of the cortisol response to stress. This, again, might have resulted in the delayed decline of intrusions and their vividness in the yohimbine condition. This mechanism would be in line with findings suggesting that glucocorticoids enhance memory consolidation of emotional stimuli when noradrenergic activity during encoding is increased (Roozendaal, Okuda, de Quervain, & McGaugh, 2006a). For example, it has been shown that memory for emotional images one week after encoding is enhanced by hydrocortisone administration in participants with increased noradrenergic activity at the time of encoding, while this is not the case for participants with hydrocortisone administration but without noradrenergic increase and participants in the placebo condition (Segal et al., 2014).

So far, there are no other experimental studies examining the effects of manipulating the noradrenergic system during encoding and consolidation of stressful or traumatic events on intrusions. However, our findings seem to be in line with data on stress and emotional memory in rodents and humans. Our results are compatible with data from animal studies suggesting enhanced fear memory after noradrenergic activation during consolidation (Dębiec, Bush, & LeDoux, 2011; Gazarini, Stern, Carobrez, & Bertoglio, 2013b). Furthermore, the consolidation of emotional events in healthy participants seems to be enhanced by stress (Segal & Cahill, 2009; Segal, Stark, Kattan, Stark, & Yassa, 2012; Smeets, Otgaar, Candel, & Wolf, 2008).

No differences in the number of intrusions and their vividness between the clonidine and placebo group were found. Therefore, noradrenergic inhibition via the α_2 receptor during trauma does not seem to influence consecutive intrusions, at least in healthy young women and in response to a relatively mild experimental stressor. Several studies examining emotional memories in humans after beta-adrenergic blockade during encoding have reported reduced memory performance for emotional material (O'Carroll, Drysdale, Cahill, Shajahan, & Ebmeier, 1999; Strange, Hurlemann, & Dolan, 2003; van Stegeren et al., 2005). Furthermore, healthy participants show impaired memory in a word list paradigm after administration of a single dose of clonidine during consolidation compared to placebo (Kuffel et al., 2014). In line with these results, a single dose of yohimbine during consolidation after a word list paradigm improves memory performance compared to placebo (Wingenfeld et al., 2013). A mechanism underlying the fact that intrusions did not differ between the clonidine and placebo group could be the activity of the HPA axis, which did not differ with respect to saliva cortisol between both groups. Furthermore, a floor effect might be responsible for the failure to find an effect of clonidine.

There are some limitations to this study. We examined intrusions in healthy participants and, therefore, it is not clear whether the data can be applied to the development of clinical PTSD. The sample was restricted to women and female sex hormones may impact intrusive memories (Ferree, Kamat, & Cahill, 2011). Therefore, the results might not be applicable to men. Also, participants taking oral contraceptives were not excluded and the cycle phase was not matched. We did, however, control for both. Furthermore, in our study design the duration of action of clonidine and yohimbine made it impossible to distinguish between noradrenergic effects during encoding or consolidation of the trauma film on the development of intrusions. This distinction might be especially important regarding future medical treatment interventions, which could more easily be administered during consolidation after the trauma, than during encoding of the traumatic event. Furthermore, the self-report of intrusive memories might be inaccurate if the participants fail to recognize the intrusions or do not report them (Takarangi, Strange, & Lindsay, 2014). However, participants' compliance was

increased by the daily transfer of recorded intrusions from the paper to the online diary and the reminder per text message.

Future research should focus on investigating the effect of the noradrenergic system and the HPA axis on the formation of intrusions separately by respectively blocking one of the systems. Furthermore, the influence of the noradrenergic system on consecutive intrusions should be evaluated in men and with other trauma film paradigms. In summary, a delayed decrease in intrusive memories as well as a delayed decrease in their vividness after yohimbine administration compared to clonidine and placebo was found. These results suggest an influence of noradrenergic activation on intrusion formation. These findings contribute to a better understanding of the neurobiological formation of intrusive memories and might contribute to the development of future intervention strategies to prevent PTSD after trauma.

4 Impact of Exogenous Cortisol on the Formation of Intrusive Memories in Healthy Women (Study II)

This chapter has been published as 'Rombold, F., Wingenfeld, K., Renneberg, B., Schwarzkopf, F., Hellmann-Regen, J., Otte, C., & Roepke, S. (2016). Impact of exogenous cortisol on the formation of intrusive memories in healthy women. *Journal of Psychiatric Research.* 83, 71-78.'

DOI: http://dx.doi.org/10.1016/j.jpsychires.2016.08.005

4.1 Abstract

Introduction: Stress hormones such as cortisol are involved in modulating emotional memory. However, little is known about the influence of cortisol on the formation of intrusive memories after a traumatic event. The aim of this study was to examine whether cortisol levels during encoding and consolidation of an intrusion-inducing trauma film paradigm would influence subsequent intrusion formation.

Methods: In an experimental, double-blind, placebo-controlled study a trauma film paradigm was used to induce intrusions in 60 healthy women. Participants received a single dose of either 20 mg hydrocortisone or placebo before watching a trauma film. Salivary cortisol and alpha-amylase as well as blood pressure were measured during the experiment. The consecutive number of intrusions, the vividness of intrusions, and the degree of distress evoked by the intrusions resulting from the trauma film were assessed throughout the following seven days.

Results: Hydrocortisone administration before the trauma film resulted in increased salivary cortisol levels but did not affect the consecutive number of intrusions, the vividness of intrusions, and the degree of distress evoked by the intrusions throughout the following week.

Conclusions: These results indicate that pharmacologically increased cortisol levels during an experimental trauma film paradigm do not influence consecutive intrusive memories. Current data do not support a prominent role of exogenous cortisol on intrusive memories, at least in healthy young women after a relatively mild trauma equivalent.

4.2 Introduction

Intrusive memories in PTSD are defined as recurrent involuntary and intrusive recollections of a traumatic event (American Psychiatric Association, 2013). Increased perceptual priming, increased associative learning, and decreased memory elaboration seem to be memory mechanisms involved in intrusion formation and intrusion maintenance (Ehlers, 2010; Wingenfeld & Wolf, 2014b). However, little is known about the neurobiological factors influencing the formation of intrusions.

During stress the HPA axis is activated and the adrenal cortex releases cortisol (de Quervain, Aerni, Schelling, & Roozendaal, 2009). Cortisol enters the brain where it binds to mineralocorticoid and glucocorticoid receptors (Wolf, Atsak, de Quervain, Roozendaal, & Wingenfeld, 2016). Hereby acting in brain regions like the amygdala, the hippocampus and the prefrontal cortex, brain regions closely associated with cognition (McEwen, Nasca, & Gray, 2016). Many studies have demonstrated that released stress hormones or exogenously administered stress hormones are involved in modulating memory consolidation (Wingenfeld & Wolf, 2014a). For example, healthy female and male participants with increased cortisol levels as a response to the Trier Social Stress Test-Modified showed enhanced memory for test day details after two weeks (Quas, Yim, Rush, & Sumaroka, 2012). Especially, cortisol enhances emotional memory consolidation (Wolf, 2009). For instance, healthy men and women who received hydrocortisone compared to placebo showed enhanced memory for emotionally arousing pictures after one week (Buchanan & Lovallo, 2001). Further, healthy men showed enhanced memory for emotional material and decreased memory for neutral material 24 hours after receiving hydrocortisone compared to placebo (Kuhlmann & Wolf, 2006). Also, the interaction of cortisol and noradrenalin seems to be involved in emotional memory formation (Roozendaal & McGaugh, 2011). For example, it has been shown that memory consolidation is enhanced by glucocorticoids when noradrenergic activity is elevated during encoding of emotional stimuli (Roozendaal et al., 2006a). Further, hydrocortisone administration before encoding of emotional images leads to enhanced memory after one week, however only in participants with increased noradrenergic activity

during encoding compared to participants without increased noradrenergic activity (Segal et al., 2014).

Several but not all studies have shown that cortisol levels after trauma are associated with subsequent intrusive symptoms and overall PTSD symptomatology in patients. For example, urinary cortisol assessed after a traumatic accident in children was associated with subsequent intrusive symptoms after six weeks (Delahanty et al., 2005). Further, women with lower serum cortisol levels measured 72 hours after sexual assault showed more overall PTSD symptoms after six weeks compared to women with higher serum cortisol levels (Walsh et al., 2013). However, plasma, saliva, and urinary cortisol after trauma assessed in the emergency room was not associated with consecutive intrusions after five months (Shalev et al., 2008). The association of cortisol levels after trauma and PTSD symptomatology seems to be complex since McFarlane and colleagues (2011) found an association of lower morning salivary cortisol and subsequent PTSD as well as higher afternoon salivary cortisol and subsequent PTSD 48 hours after trauma.

Patients who received hydrocortisone during intensive care unit (ICU) treatment showed reduced PTSD symptoms (Schelling et al., 2006). For example, compared to placebo, hydrocortisone administration during septic shock (Schelling et al., 2001; Schelling et al., 1999) and during cardiac surgery (Schelling et al., 2004) was associated with a decreased incidence of PTSD (Schelling et al., 2001; Schelling et al., 1999) and reduced PTSD symptoms (Schelling et al., 2004). In a recent meta-analysis hydrocortisone was the only early pharmacological intervention to effectively prevent PTSD (Sijbrandij et al., 2015). For instance, female and male patients with heterogeneous injuries after a traumatic event who received either placebo or 20 mg hydrocortisone within 12 hours post-trauma and subsequently every 12 hours for the following ten days reported less PTSD symptoms during the three months post-trauma compared to placebo (Delahanty et al., 2013). Further, a reduced risk for subsequent PTSD three months after trauma was found in female and male trauma survivors who received intravenous hydrocortisone six hours after trauma (work or vehicle accidents) compared to placebo (Zohar et al., 2011). PTSD patients show decreased basal cortisol levels in most but not all studies (Yehuda et al., 2015). Exogenous hydrocortisone in PTSD

patients inhibited retrieval of trauma related memories in a case series of patients (Aerni et al., 2004), however, in a recent study no effect of exogenous hydrocortisone on intrusive memories in female PTSD patients emerged (Ludäscher et al., 2015).

Trauma film paradigms are used to examine the formation of intrusive memories by inducing short lasting intrusions in healthy participants (Holmes & Bourne, 2008). First evidence suggests that cortisol also influences intrusion formation. In a study in healthy participants using a trauma film paradigm, there was a positive correlation between post-film salivary cortisol levels and the frequency of intrusions, however this was only the case in participants with increased sAA activity (Chou et al., 2014), an indicator of enhanced noradrenergic activation (van Stegeren et al., 2006).

The present study examined the potential influence of exogenous cortisol during a trauma film paradigm on the consecutive development of intrusions. Based on the available data on emotional memory consolidation and intrusion formation, we hypothesized that an increased number of consecutive intrusions, more vivid intrusions, and more distressing intrusive memories of the trauma film would be found in participants who received hydrocortisone compared to participants who received placebo.

4.3 Methods

4.3.1 Participants

The sample consisted of 60 healthy university students. Recruitment took place via flyers posted around the university, official university email lists, or postings in social network university groups. As the rape victim in the trauma film is female, only women were included. The SCID-I (First et al., 1995) was conducted over the phone to exclude participants with former or present DSM-IV Axis I disorders. Furthermore, physical illnesses, medication intake (except oral contraceptive), history of sexual abuse or rape, pregnancy, and lactation period led to exclusion.

The HCG ULTRA pregnancy test was implemented to ensure the exclusion of pregnant woman. All participants spoke German on a native level and were between 18 and 34 years old (see Table 2).

Table 2. *Sample characteristics*

Characteristic	Hydrocortisone *n* = 30 **M(SD) or n(%)**	Placebo *n* = 30 **M(SD) or n(%)**	Statistics	*p*
Age	23(3.32)	22.7(3.41)	$t(58) = .35$	.73
Intake of oral contraceptives	10(33.3 %)	14(46.7 %)	χ^2 (1) = 1.1	.29
Current smoker	6(20.0 %)	4(13.3 %)	χ^2 (1) = .48	.49
Menstrual cycle, follicular vs. luteal phase	17(56.7 %) 13(43.3 %)	14(48.3 %) 15(51.7 %)	χ^2 (1) = .42	.52
BMI	22.04(2.7)	21.96(2.89)	$t(58) = .11$	.92
CTQ	36.83(6.98)	40.77(9.78)	*t(58) = -1.8*	.08
BDI	4.73(5.1)	5(3.23)	*t(58) = -.21*	.83
STAI-T	34.78(8.84)	35.27(6.7)	*t(58)* = -.25	.81

Abbreviations: BMI, Body Mass Index; CTQ, Childhood Trauma Questionnaire; BDI, Beck Depression Inventory; STAI-T, trait scale of the Trait Anxiety Inventory.

4.3.2 Procedure

A double-blind, randomized, placebo-controlled study design was applied. The study was conducted at the Department of Psychiatry and Psychotherapy, Campus Benjamin Franklin, Charité - Universitätsmedizin Berlin. All procedures were approved by the local ethics committee of the German Psychology Association (DGPs) and participants provided written informed consent at least 24 hours in advance. Participants were instructed to refrain from smoking, physical exercise, consuming caffeine, drinking alcohol or other beverages (except for water), and

eating at least one hour prior to the assessment. One participant was tested per day at 1.30 pm.

Experimental phase

Participants received either hydrocortisone (two tablets of 10 mg, Galen ®) or placebo (two tablets of Placebo, Lichtenstein®). They were randomly allocated to one of the groups. The hydrocortisone and placebo pill looked identical. The investigator was excluded from the randomization protocol, hence ensuring that the participant and the investigator were blind to the treatment condition. Hydrocortisone or placebo was administered sixty minutes prior to the trauma film. The time of administration and the dosage have been successfully deployed in a previous study (Buchanan & Lovallo, 2001). Saliva was collected and blood pressure was measured (automatic device: boso medicus uno, Bosch + Sohn Germany) seven times throughout the study: at baseline, after medication intake, and every 15 minutes five times after the trauma film. Childhood trauma has been associated with increased risk to develop PTSD after trauma in adulthood (Breslau et al., 1999). To control for differences in experienced childhood events the CTQ (Bernstein & Fink, 1998; Wingenfeld et al., 2010) was applied. It has been shown that pre-trauma trait anxiety is positively related to PTSD (McNally et al., 2011). Therefore, trait anxiety was measured with the STAI-T (Laux et al., 1981; Spielberger & Gorsuch, 1983). Participants were instructed to not talk to other potential participants about the content of the study. Participants filled out the intrusion diary for seven days subsequent to watching the trauma film.

Follow-up session

Participants returned the diary on the eighth day after the trauma film. The IES (Horowitz et al., 1979) was applied to evaluate intrusion, avoidance, and hyperarousal as PTSD related symptoms. Via phone the IES was conducted again after four weeks and debriefing was provided in oral and written form at the end. Participants received a 35 Euro incentive.

4.3.3 Trauma film and intrusion diary

To evoke short lasting intrusions, a trauma film paradigm which has been successfully used in other studies was adopted (Weidmann et al., 2009). The scene from the film '*Irréversible*' (Gaspar Noé, 2002; 14 minutes 40 seconds) contains severe sexual and physical violence, depicting the rape of a woman by a man in detail and has been shown to induce stress and intrusions (Weidmann et al., 2009). The film was shown on a 2 x 2.5 meter screen while the sound was played through headphones. To ensure that all participants watched the film without closing their eyes or taking off the headphones, the female researcher was present while the participant watched the film.

Participants were instructed to record their intrusions in a pencil and paper diary (Holmes et al., 2004; Weidmann et al., 2009) throughout the following seven days. In addition, the notes from the pencil and paper diary had to be transferred to an identical online diary, allowing the instructor to check on participation. To remind participants to transfer intrusions to the online diary, a text message was sent at 9 p.m. daily. The difference between image and thought based memories was explained to the participant followed by this instruction: "record any spontaneously occurring memory from the trauma film instantaneously after occurrence". Additionally, memories had to be specified by spontaneity of appearance, frequency, modality (image, thought based, or both), content, vividness, and the degree of distress (Holmes et al., 2004). The participant rated the vividness and the degree of distress of every intrusive memory from zero ("not at all") to five ("a lot"). The instructor evaluated the vividness and the degree of distress by summing up daily ratings. Memories were considered intrusions and included in the analyses if they met the following criteria (Arntz et al., 2005; Ehlers et al., 2004): (1) imagery based, (2) vividness > 0, (3) degree of distress > 0, (4) spontaneously occurring. To remove any doubts about the inscriptions in the diary, the female researcher went through each memory with the participant and rated the memory to be intrusive or not intrusive.

4.3.4 Salivary assessment

Saliva was collected using salivettes (Salivettes®, blue cap; Sarstedt) and stored after the assessment at -20 °C. Homogenous saliva collection from all salivary glands was ensured by the following instruction: "move the cotton swab in your mouth in a circular pattern for one minute without actively chewing on it" (Nater & Rohleder, 2009). Both, salivary free cortisol and sAA were determined by the Neurobiology Laboratory of the Department of Psychiatry and Psychotherapy, Charité - Universitätsmedizin Berlin. Inter- and intra-assay coefficients of variation were both below 12 % for salivary cortisol and both lower than 10 % for sAA activity.

4.3.5 Statistical analysis

Differences between treatment groups in clinical or demographic variables were determined by independent Student's t-tests for continuous data and Pearson's chi-squared test for categorical data. Both for total cortisol and total sAA, the area under the curve (AUC) was calculated (Pruessner, Kirschbaum, Meinlschmid, & Hellhammer, 2003). Pearson's correlation analyses were performed to analyze a possible association between salivary cortisol (AUC), sAA (AUC), number of intrusions, vividness of intrusions, and degree of distress evoked by the intrusions. Further, a possible association between the menstrual cycle phase and the number of intrusions, the vividness of intrusions and the degree of distress evoked by the intrusions was analyzed. To analyze the effects of treatment (between-subjects factor) and time (within-subjects variable with seven levels) on salivary cortisol levels, sAA, and blood pressure, three sets of repeated measures mixed design ANOVAs were conducted. Furthermore, the effects of treatment (between-subjects factor) and time (within-subjects variable with four levels) on number of intrusions, vividness, and degree of distress as dependent variables were examined by ANOVA. For repeated-measures ANOVA, η_p^2 was used as effect size. Homogeneity of variance was assessed by Levene's statistic and sphericity was examined with Mauchly's test in all ANOVAs. Given the case that the assumption of sphericity was not met, Greenhouse–Geisser corrected p values are reported.

Bonferroni correction for multiple comparisons was performed for post-hoc comparisons. Statistical analyses were performed using SPSS Version 23.0 (SPSS, Chicago, IL, USA). The alpha level for significance was set to 0.05.

4.4 Results

4.4.1 Participant characteristics

Demographic variables, possible cofounding variables, and related statistics are presented in Table 2. Participants in the hydrocortisone and placebo group did not significantly differ in any variable.

4.4.2 Salivary cortisol

Figure 8 displays salivary cortisol levels throughout the assessment. Both, the effect of time ($F_{\text{df2.52,145.96}}$ = 29.24, p < .01, η_p^2 = .34) and the effect of treatment ($F_{\text{df1,58}}$ = 92.19, p < .01, η_p^2 = .61) were significant. Furthermore, the time x treatment interaction was significant ($F_{\text{df2.52,145.96}}$ = 29.72, p < .01, η_p^2 = .34). Post-hoc tests revealed higher salivary cortisol levels in the hydrocortisone group compared to the placebo group. There was no difference in salivary cortisol levels between the two groups at baseline (p > .05).

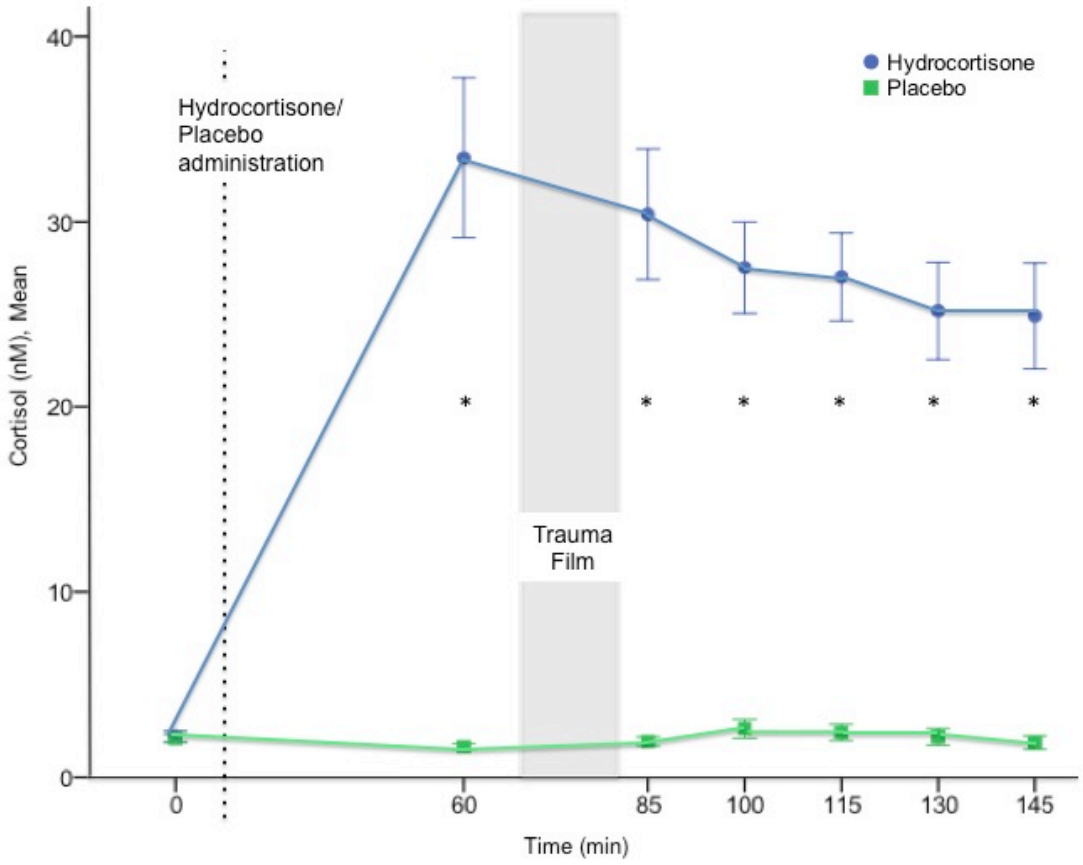

Figure 8. Salivary Cortisol levels at baseline, before, and after the trauma film for the hydrocortisone and placebo group. Error bars represent +/- 1 SE.

4.4.3 Salivary alpha-amylase and blood pressure

Figure 9 displays the sAA activity for the two treatment groups in the course of the assessment. A significant effect of time ($F_{df3.43,185.14} = 12.28$, $p < .01$, $\eta_p^2 = .19$) was revealed. Neither the effect of treatment ($F_{df1,54} = 1.83$, $p = .18$) nor the time x treatment interaction ($F_{df3.43,185.14} = .17$, $p = .93$) was significant.

The effect of time for diastolic ($F_{df4.39,254.77} = 7.98$, $p < .01$, $\eta_p^2 = .12$) and systolic ($F_{df4.71,273.12} = 8.14$, $p < .01$, $\eta_p^2 = .12$) blood pressure was significant. There was no significant effect of treatment for diastolic ($F_{df1,58} = 3.81$, $p = .06$) and systolic ($F_{df1,58} = 1.39$, $p = .24$) blood pressure. There was no significant interaction effect time x treatment for both diastolic ($F_{df4.39,254.77} = 1.01$, $p = .41$) and systolic blood pressure ($F_{df4.71,273.12} = 1.77$, $p = .12$).

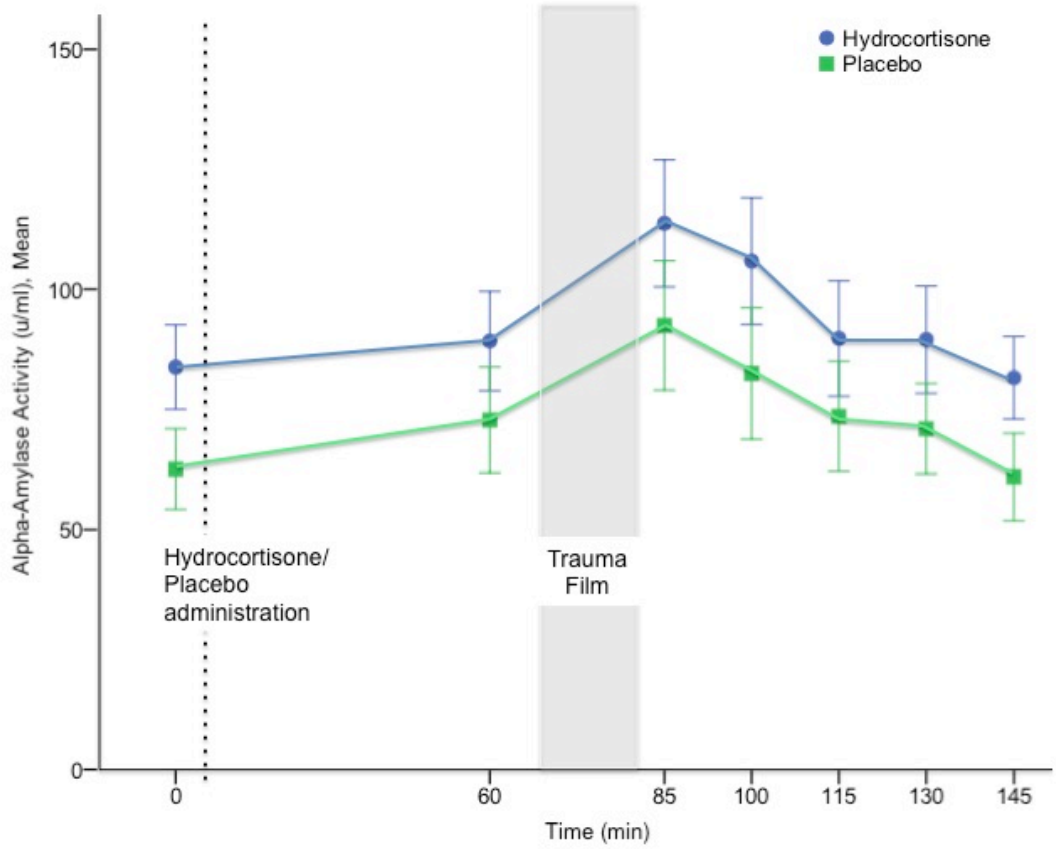

Figure 9. Salivary alpha-amylase activity at baseline, before, and after the trauma film for the hydrocortisone and placebo group (* $p < .05$). Error bars represent +/- 1 SE.

4.4.4 Intrusions

Figure 10 displays the number of intrusions over seven days for both groups. A significant effect of time ($F_{df2.09,121.39} = 22.43$, $p < .01$, $\eta_p^2 = .28$) indicates a decline of intrusions over the course of seven days. No significant effect of treatment ($F_{df1,58} = .00$, $p = .99$, $\eta_p^2 < .01$) and no significant time x treatment interaction ($F_{df2.09,121.39} = .73$, $p = .49$, $\eta_p^2 = .01$) was observed. Only five participants did not report any intrusions.

Figure 11 displays the vividness of intrusions over seven days for both treatment groups. The vividness of intrusive memories declined over time ($F_{df1.7,98.45} = 20.69$, $p < .01$, $\eta_p^2 = .26$). Both, the effect of treatment ($F_{df1,58} = .06$, $p =$

.82, η_p^2 < .01) and the time x treatment interaction ($F_{df1.7,98.45}$ = .84, p = .42, η_p^2 = .01) were not significant.

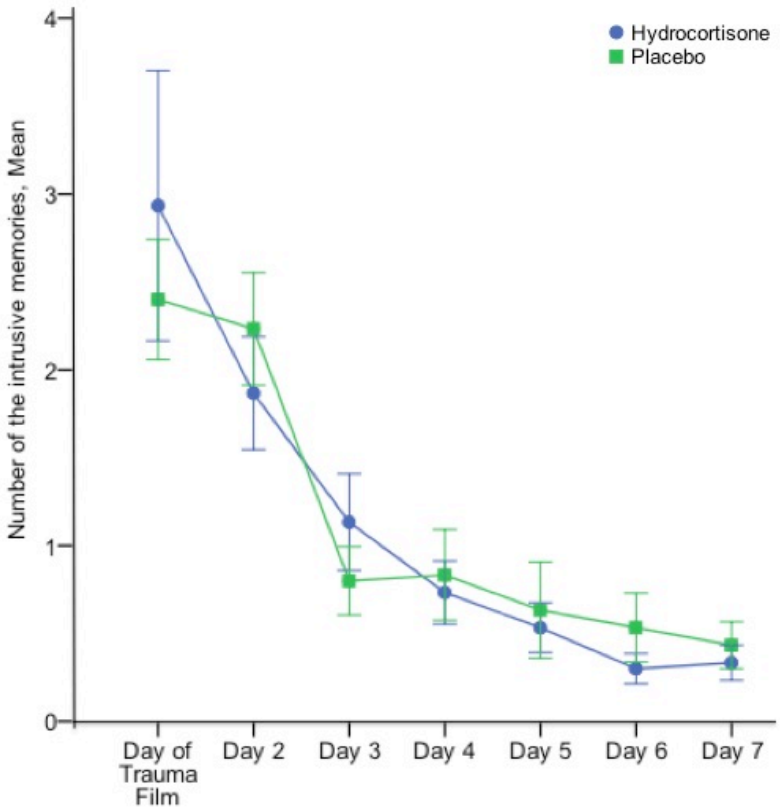

Figure 10. Results for the number of intrusions for the hydrocortisone and placebo group over seven days, starting on the day of the trauma film. Error bars represent +/- 1 SE.

Figure 12 displays the degree of distress participants in both groups experienced because of the intrusions throughout the seven days. There was a significant effect of time ($F_{df1.94,112.6}$ = 24.16, p < .01, η_p^2 = .29) indicating a decline of distress over the course of seven days. There was no significant effect of treatment ($F_{df1,58}$ < .01, p = .97, η_p^2 < .01) and no treatment x time interaction, $F_{df1.94,112.6}$ = .69, p = .5, η_p^2 = .01).

Including sAA activity (AUC) or intake of oral contraceptives separately as covariates in the general linear model did not modify the results, no significant effect of treatment group emerged in all three analyses (number of intrusions, vividness, and degree of distress).

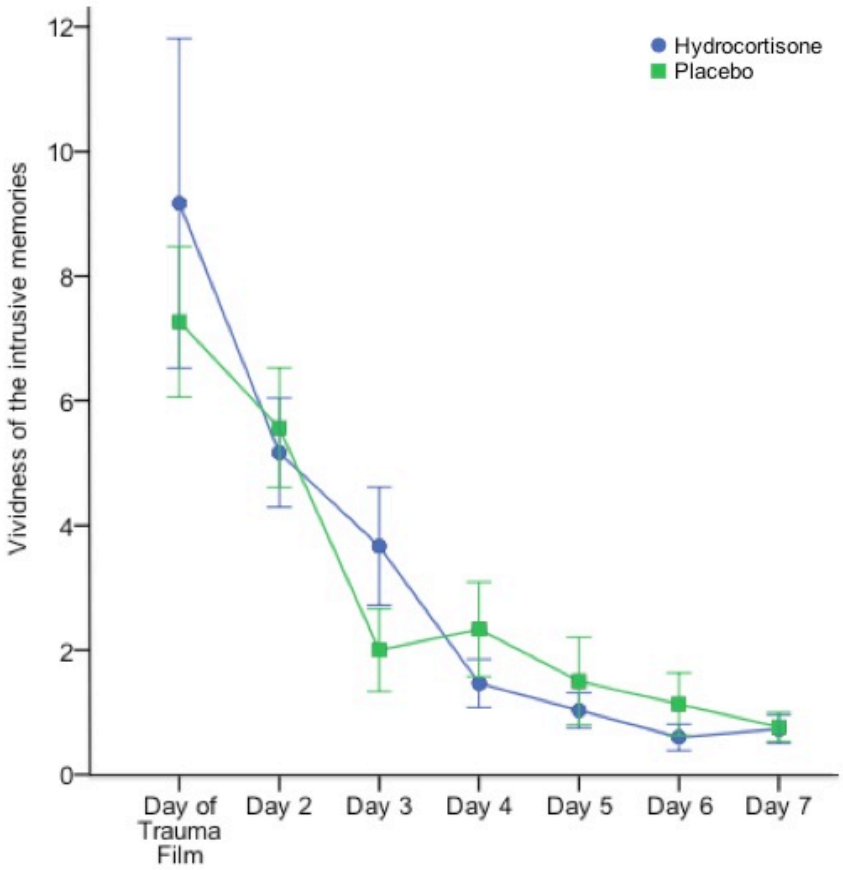

Figure 11. Results for the vividness of intrusions for the hydrocortisone and placebo group over seven days, starting on the day when the trauma film. Error bars represent +/- 1 SE.

In the hydrocortisone group the cortisol level (AUC) was associated with the number of intrusions on day two (r = .42, p = .02), however no further correlations between salivary cortisol (AUC), sAA (AUC), number of intrusions, vividness of intrusions, and degree of distress evoked by the intrusions were found. The menstrual cycle phase was not associated with the number of intrusions (r = .03), the vividness of intrusions (r = -.01) or the degree of distress (r = -.03) evoked by the intrusions (all p > .94).

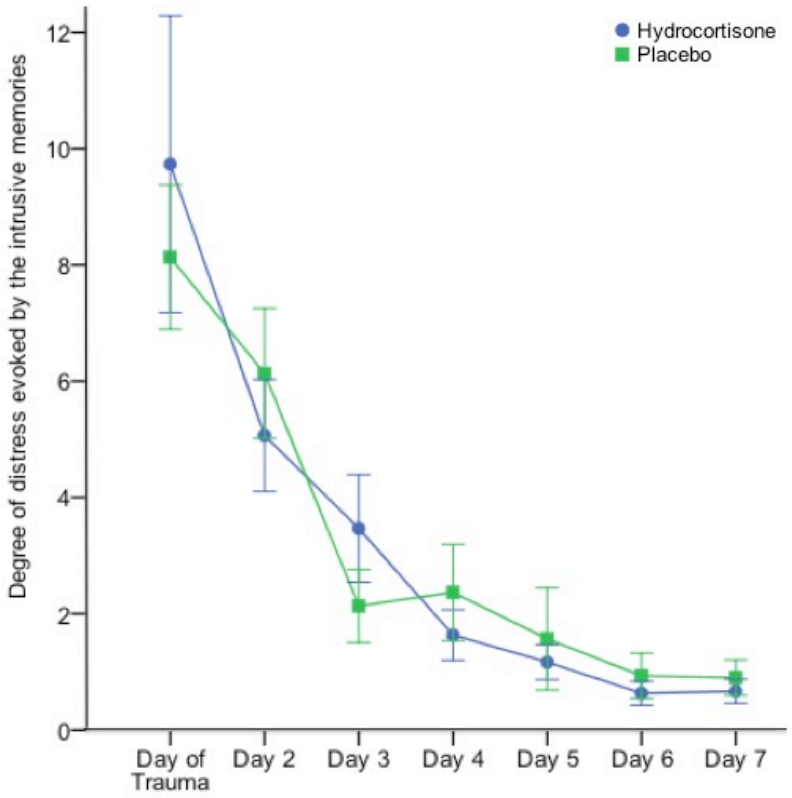

Figure 12. Results for the degree of distress evoked by the intrusions for the hydrocortisone and placebo group over seven days, starting on the day when the trauma film. Error bars represent +/- 1 SE.

4.4.5 The Impact of Event Questionnaire

The hydrocortisone and placebo group did not differ in PTSD related symptoms assessed with the IES (Horowitz et al., 1979; Maercker & Schützwohl, 1998), neither one week (t_{df58} = .33, p = .74) nor four weeks (t_{df58} = .16, p = .88) after the assessment.

4.5 Conclusions

This study examined the influence of pharmacologically increased cortisol levels during encoding and consolidation of a trauma film on the consecutive formation of intrusive memories in healthy women. The number of intrusions, their vividness, and the degree of distress evoked by the intrusions throughout the

course of one week after the trauma film did not differ between the hydrocortisone and the placebo group.

The results of the current study indicate that pharmacologically increased cortisol levels during an experimentally induced trauma do not have an impact on consecutive intrusive memories of the trauma event in healthy women, also when endogenous sAA activity and therefore noradrenergic activation is included in the analysis. The results do not concur with the assumption that intrusive memory formation relies on a similar biological mechanism as emotional memory consolidation, where cortisol levels are positively related to emotional memory formation (Roozendaal & McGaugh, 2011). For example, exogenous cortisol during consolidation increased emotional memory in healthy controls (Buchanan & Lovallo, 2001; Kuhlmann & Wolf, 2006). Further, the corticosteroid synthesis inhibitor metyrapone administered before TSST impaired memory consolidation of a following picture story compared to placebo and propranolol in healthy men (Maheu, Joober, Beaulieu, & Lupien, 2004). Additionally, healthy women who received a single dose of hydrocortisone prior to viewing neutral and emotional pictures displayed emotional memory enhancement after one week, but this was only the case if they additionally showed an increase in endogenous sAA during consolidation and not only with elevated cortisol levels (Segal et al., 2014).

First empirical evidence on the influence of endogenous cortisol on intrusion formation has been contradictory (Bryant et al., 2013; Chou et al., 2014; Nicholson et al., 2014). Participants with increased endogenous sAA activity during a trauma film paradigm showed a positive correlation between post-film endogenous salivary cortisol levels and the frequency of intrusions (Chou et al., 2014). The results of the present study, even after taking endogenous sAA activity into account, do not support this finding of an involvement of the HPA axis in intrusion formation. However, our findings extend the results of a study where intrusive memories of negatively valenced pictures were measured and a positive synergistic effect of noradrenergic activation and cortisol on intrusive memory formation was replicated only in men but not in women (Bryant et al., 2013). Furthermore, in accordance with our data, endogenous cortisol was not associated with intrusion formation after seeing negatively valenced pictures in healthy participants, only in PTSD patients with increased endogenous sAA (Nicholson et

al., 2014). Of note, in the last two studies (Bryant et al., 2013; Nicholson et al., 2014) intrusive memories were only assessed retrospectively after two days in comparison to a diary assessment over seven days applied in the current study.

Hydrocortisone administration 60 minutes prior to a trauma film did not influence consecutive intrusion formation in the current study. However, recent research indicates a time dependent effect of cortisol on memory formation (van Ast, Cornelisse, Meeter, Joëls, & Kindt, 2013). Non-genomic effects appear first after cortisol administration followed by the slower genomic effects starting 60 minutes after cortisol exposure (Cornelisse, van Ast, Joëls, & Kindt, 2014). Empirical data show that trace conditioned memory is enhanced when cortisol is administered 240 minutes (genomic effects) prior fear acquisition, however, this is not the case if cortisol is administered 60 minutes (non-genomic effects) prior fear acquisition (Cornelisse et al., 2014). Therefore, the possibility cannot be ruled out that later genomic effects of cortisol might be involved in intrusion formation.

The strength of this study lies in the paradigm that is able to induce intrusions within a randomized controlled design. However, some limitations should be recognized. Intrusive memories were examined in healthy participants following a relatively mild stressor compared to real traumatic events that can cause PTSD. Therefore, it is uncertain whether current results can also account for the formation of intrusive memories in patients with PTSD. Further, vulnerable populations, such as individuals with previous traumatic experiences, might respond differently to cortisol during encoding and consolidation of traumatic events. Healthy non-traumatized young women might be less suitable to study the impact of cortisol on intrusion formation. Furthermore, emotional memory consolidation differs between men and women (Felmingham et al., 2012) and female sex hormones impact the formation of intrusions (Ferree et al., 2011). For example, salivary estrogen in women is associated with increased intrusions (Cheung, Chervonsky, Felmingham, & Bryant, 2013). Therefore, the results from a female sample might not be transferable to men. Emotional memory consolidation also differs between women with a natural cycle and women taking oral contraception (Nielsen, Barber, Chai, Clewett, & Mather, 2015). Therefore, we controlled for oral contraceptive intake. In addition, the duration of action and the time of administration of hydrocortisone do not allow the discrimination between

the effects of hydrocortisone during encoding or consolidation of the trauma film on the formation of intrusive memories. The lack of influence of exogenous cortisol on intrusion formation in the current study might also be explained by the sensitivity of the task. The chosen method to record the intrusive memories relies on self-report and it has been shown that participants do not always recognize their intrusions and do sometimes not report them (Takarangi et al., 2014). However, to date there is no other method known to the authors to examine individual human intrusive memories. The instructed daily transfer of intrusions from the paper diary to the online diary and the daily text message increased compliance. Further, a floor effect due to the overall small number of intrusions could be responsible for the negative finding: a lack of influence of exogenous cortisol on intrusion formation. Lack of power seems unlikely, as the effect size of effect of treatment (and treatment by time interaction) on intrusions was shown to be very small.

Future research should focus on the time dependent effects of cortisol and study late, genomic effects of cortisol on intrusion formation. Furthermore, the endocannabinoid system seems to be involved in emotional memory consolidation (Wolf et al., 2016) and its influence in intrusion formation should be considered in future research. Additionally, genetic and epigenetic mechanisms within the HPA axis might be involved in intrusion formation (Cheung & Bryant, 2015) and should be studied. Finally, the interactional influence of the HPA axis and noradrenergic systems should be examined by blocking/ activating one system while activating/ blocking the other system.

To conclude, no differences in the number of intrusions, their vividness, and the degree of distress evoked by the intrusions between healthy women receiving either hydrocortisone or placebo before the trauma film paradigm were found. The results provide preliminary evidence that high cortisol levels after exogenous hydrocortisone administration during trauma has no strong impact on consecutive intrusion formation.

5 Impact of Stress Response Systems on Forced Choice Recognition in an Experimental Trauma Film Paradigm (Study III)

This chapter will be published as 'Rombold-Bruehl, F., Otte, C., Renneberg, B., Bruch, L., Wingenfeld, K., & Roepke, S. (2017). Impact of stress response systems on forced choice recognition in an experimental trauma film paradigm. Manuscript submitted for publication.'

5.1 Abstract

Introduction: Traumatic events are often followed by memory impairments of key features of the trauma. Stress hormones are involved in emotional memory formation. However, little is known about their influence during trauma on subsequent recognition of trauma details.

Methods: A pooled analysis of two double-blind, placebo-controlled studies (N = 175) was performed to assess the influence of the noradrenergic system and the HPA axis on intrusion formation. Participants received either 10 mg yohimbine (stimulating noradrenergic activity), 0.15 mg clonidine (inhibiting noradrenergic activity), or placebo (study one) or 20 mg hydrocortisone or placebo (study two), each 60 minutes before watching a distressing film depicting severe sexual and physical violence. After seven days, the participants performed a 24-item forced choice recognition test. Memory was assessed for pre-, peri-, and post-trauma film scenes.

Results: A significant film scene by intervention interaction indicated a differential influence of drug intervention on the number of correct pre-, peri-, and post-trauma film scene memories one week after the distressing film. Post-hoc tests revealed that clonidine led to significantly fewer correct peri-trauma film scene memories compared to placebo and, on a trend level, to yohimbine.

Conclusions: Pharmacological inhibition of noradrenaline during a distressing film leads to impaired emotional memory recognition for peri-trauma film scene details.

5.2 Introduction

Hormones released during stressful events influence learning and memory in healthy humans (Roozendaal & McGaugh, 2011). While emotional life events are better remembered than emotionally non-arousing events (McGaugh, 2003), stress can impair memory retrieval (de Quervain et al, 2009). Stress hormones released by the noradrenergic system and the HPA axis are involved in emotional memory formation in healthy participants (Brewin et al., 1996; Roozendaal & McGaugh, 2011; Schwabe et al., 2012; van Stegeren, 2008). However, the influence of stress hormones released by the noradrenergic system and the HPA axis during a traumatic event on subsequent memory of the respective event needs to be examined.

The activity of the noradrenergic system during encoding and consolidation influences the formation of emotional memory as shown by a large number of studies (Roozendaal & McGaugh, 2011; Schwabe et al., 2012; van Stegeren, 2008). Animal studies show that noradrenergic activation of the basolateral nucleus of the amygdala during stress (McGaugh & Roozendaal, 2009) seems to be responsible for the strengthening of consolidation processes in the hippocampus (McGaugh, 2004). For example, within a Pavlovian threat fear conditioning paradigm, activation of β-adrenergic receptors through noradrenaline infusion into the lateral nucleus of the amygdala leads to enhanced freezing during the paradigm (Schiff et al, 2016). The majority of studies with healthy participants shows that pharmacologically increased noradrenergic activity with an α2-adrenoceptor antagonist (e.g., yohimbine) during encoding and consolidation leads to enhanced emotional memory formation, whereas blocking of the adrenergic receptor with a β-adrenergic antagonist (e.g., propranolol) leads to impaired emotional memory formation (Chamberlain & Robbins, 2013). For example, Cahill and colleagues (1994) administered either propranolol or placebo one h before healthy participants listened to an emotional or neutral narrative accompanied by a series of slides. These authors found selective impairing effects of propranolol on memory of the presented emotional story one week later. Further, healthy participants who received clonidine, an α2-adrenergic agonist inhibiting noradrenergic activity, before a word list learning paradigm, showed impaired

memory of the word list 24 hours after encoding (Kuffel et al., 2014). Additionally, in healthy participants who had been exposed to emotionally arousing pictures increased sAA after exposition, an indicator of noradrenergic activity, was positively associated with the percentage of remembered pictures after one week (Segal & Cahill, 2009).

The HPA axis releases cortisol during stress (Dominique, Aerni, Schelling, & Roozendaal, 2009) which binds to mineralocorticoid and glucocorticoid receptors (Wolf et al., 2016) located in the amygdala, the prefrontal cortex, and the hippocampus (McEwen et al., 2016). The HPA axis also influences emotional memory (Finsterwald & Alberini, 2014; Schwabe et al., 2012; Wolf, 2009). For example, increased cortisol levels in young, healthy participants following the Trier Social Stress Test-Modified have been associated with more remembered details of the test day after two weeks (Quas et al., 2012). Further, the administration of hydrocortisone before viewing emotional and neutral pictures resulted in enhanced emotional memory after one week in healthy women with increased endogenous sAA during consolidation (Segal et al., 2014). Additionally, following the administration of hydrocortisone, healthy participants showed improved memory for emotionally arousing material compared to neutral material (Kuhlmann & Wolf, 2006).

To date it is unknown whether the activity of the noradrenergic system and the HPA axis during trauma influences subsequent memory for trauma details. Intentional recall deficits of trauma-details six months (Halligan, Michael, Clark, & Ehlers, 2003) and one year (Rothbaum, Foa, Riggs, Murdock, & Walsh, 1992) after the event have been associated with PTSD diagnosis in assault victims. PTSD patients show greater disorganization in trauma memories compared to unpleasant but nontraumatic memories (Brewin et al, 2014). In previous studies we have been able to show that the noradrenergic system but not the HPA axis impacts on the decline of intrusive memories following a trauma film (Rombold et al., 2016a; Rombold et al., 2016b). However, the influence of the stress response systems during encoding and consolidation of traumatic events on recognition of trauma details is still unclear.

The trauma film paradigm has been widely used to assess the influence of trauma processing on memory for trauma film details (Rombold et al., 2016a;

Rombold et al., 2016b). A sequence from the film '*Irréversible*' (Gaspar Noé, 2002) depicting severe sexual and physical violence has been shown to induce intrusions (Weidmann et al, 2009).

In this study we assessed the influence of pharmacologically increased and decreased noradrenergic activity as well as exogenous cortisol administration during a distressing trauma film on memory for film details after one week in healthy women. To study differential effects of the stress response systems on film scenes with or without traumatic content, we compared pre-trauma, peri-trauma (i.e., sexual and physical assault), and post-trauma film scenes. Following data on the impact of biological stress response systems on emotional learning, we hypothesized that noradrenergic activation (i.e., yohimbine) and increased cortisol levels (i.e., hydrocortisone) would enhance memory for peri-trauma film scene details, whereas noradrenergic blockade (i.e., clonidine) would decrease memory for peri-trauma film scene details compared to placebo.

5.3 Methods

5.3.1 Participants

A pooled analysis of two studies with an identical design and healthy, female university student participants (N=178) was performed (Rombold et al., 2016a; Rombold et al., 2016b). Study one (n=118) assessed the impact of noradrenergic stimulation/blockade on forced choice recognition of distressing film details. Study two (n=60) assessed the impact of exogenous cortisol on forced choice recognition of trauma film details. The placebo groups of both studies were combined. Four participants were excluded from study one due to fatigue after clonidine intake (n=1), withdrawal while watching the film (n=2), and missing data (n=1). Recruitment took place via official university email lists and public postings. The sample was restricted to women because the victim in the film is female.

History of abuse or rape, physical illnesses, present or former DSM-IV Axis I disorders assessed by the SCID-I (First et al., 1995) , medication intake (except oral contraceptive), and pregnancy or lactation period led to exclusion. All participants

were between 18 and 44 years old (see Table 3), spoke German on a native level, and received 35 Euro for participation.

Table 3. *Sample characteristics*

Characteristic	Clonidine *n* = 38 **M(SD) or n(%)**	Yohimbine *n* = 38 **M(SD) or n(%)**	Hydro-cortisone *n* = 30 **M(SD) or n(%)**	Placebo *n* = 68 **M(SD) or n(%)**	Statistics	*p*
Age	23.3(3.6)	23.4(4.5)	23.0(3.3)	22.9(3.3)	$F_{df3,170}$ = .19	.91
Intake of oral contraceptives	19(50.0 %)	13(34.2 %)	10(33.3 %)	36(52.9 %)	χ^2 (3) = 5.56	.14
Menstrual cycle, follicular vs. luteal phase	18(50 %) 18(50 %)	20(58.8 %) 14(41.2 %)	17(56.7 %) 13(43.3 %)	32(47.8 %) 35(52.2 %)	χ^2 (3) = 1.44	.7
BMI	21.2(2.6)	21.8(3.0)	22.0(2.7)	22.2(3.0)	$F_{df3,169}$ = .99	.4
CTQ	37.4(9.1)	36.1(6.1)	36.8(6.98)	39.4(9.9)	$F_{df3,170}$ = .23	.23
DSS-Acute	1.6(1.0)	1.9(1.6)	.9(1.4)	1.6(1.4)	$F_{df3,170}$ = 3.25	.02

Abbreviations: BMI, Body Mass Index; CTQ, Childhood Trauma Questionnaire; DSS-Acute, Dissociation-Tension-Scale acute.

5.3.2 Procedure

Approval was given by the local ethics committee of the German Psychology Association (DGPs). All participants provided written informed consent at least 24 hours before the assessment. One participant was tested per day.

Experimental phase

Participants received either clonidine (0.15 mg, inhibiting noradrenergic activity, study one), yohimbine (10 mg, stimulating noradrenergic activity, study one), two tablets of hydrocortisone, a synthetic glucocorticoid (10 mg, Galen ®, study two), or placebo (Lichtenstein®, studies one and two). In each study participants were

randomly allocated to the groups. The verum and the placebo looked identical to ensure the double blind design. Considering the peak levels, administration of hydrocortisone (Buchanan & Lovallo, 2001), clonidine (Reid, 1981), and yohimbine (Owen et al., 1987), as well as administration of placebo took place 60 minutes prior to the film.

As childhood trauma has been associated with an increased neuroendocrine response to stress (Heim & Binder, 2012), the CTQ (Bernstein & Fink, 1998; Wingenfeld et al., 2010) was utilized to control for differences in experienced childhood events. The DSS-Acute (Stiglmayr et al., 2003) was applied since dissociation during trauma has been associated with disorganized and disintegrated recall of trauma details (Bedard-Gilligan & Zoellner, 2012). All participants received the instruction not to talk about the study to other potential participants.

Follow-up session

After seven days, the participants came back to the laboratory and filled out a 24-item forced choice recognition test that assessed memory of the film. The test included four right and four wrong statements for each of the pre-, peri-, and post-trauma film scene details (see Table 4). Participants had to decide if each of the 24 statements about the film was right or wrong. At the end of the study participants received a debriefing via phone and in written form.

Table 4. *Recognition test*

Item	**Film Scene**	**Right vs. Wrong**
"The woman is wearing a dress."	pre	right
"The woman has a purse."	pre	right
"The woman is flagging a taxi."	pre	right
"Prostitutes are standing next to the street."	pre	right
"The woman has blond hair."	pre	wrong
"The woman is carrying a basket."	pre	wrong
"It is a single track road but there is heavy traffic."	pre	wrong
"The woman is in a club."	pre	wrong
"The man is wearing a suit."	per	right
"The walls are red."	peri	right
"A second man appears in the background."	peri	right
"The man is wearing a ring."	peri	right
"The man has a pistol."	peri	wrong
"The man is wearing a tie."	peri	wrong
"The man has a briefcase."	peri	wrong
"Nobody witnesses the rape."	peri	wrong
"The woman lies on a stretcher."	post	right
"The friend who wanted to walk her home is leaving the party as the ambulance arrives."	post	right
"One of the men smokes while leaving the party."	post	right
"The crime scene is cordoned off."	post	right
"Leaving the party, one of the men holds a bottle of beer in his hand."	post	wrong
"The boyfriend leaves the party with a group of men."	post	wrong
"When the men leave the party the sun has already risen."	post	wrong
"The friends are stopped by an ambulance man."	post	wrong

5.3.3 Trauma film

A scene (14 minutes, 40 seconds) from the film '*Irréversible*' (Gaspar Noé, 2002) with severe sexual and physical violence in which a women is raped by a man on her way home was shown to all participants on a 2 x 2.5 meter screen. The female researcher was present in the darkened room to ensure that the participant watched and listened to the film. The film scene was categorized into pre-, peri-, and post-trauma film scenes. During the pre-trauma film scene the woman attends a party, during the peri-trauma film scene the women is anally raped and beaten by a man, and during the post-trauma film scene an ambulance car is arriving to rescue the severely injured woman.

5.3.4 Statistical analysis

Statistical analyses were performed with SPSS Version 23.0. Statistical significance refers to a p value < 0.05 and an effect on a trend level refers to a p value < 0.1.

Demographic data and potential differences between intervention groups in the conducted questionnaires were analyzed using Pearson's chi-squared test for categorical data and univariate ANOVA for continuous data. Repeated measures mixed design ANOVA was used to examine the effects of intervention (between-subjects factor) and film sequence (within-subjects variable with three levels) on number of correct trauma film specific memories as the dependent variable.

Homogeneity of variance was assessed by Levene's statistic and sphericity was examined with Mauchly's test. For post-hoc comparisons, a Bonferroni correction for multiple comparisons was performed.

5.4 Results

5.4.1 Participants Characteristics

The yohimbine, clonidine, hydrocortisone, and placebo group did not differ significantly in any demographic variable or possible cofounding variable except in

dissociation during the film which was therefore included in the analysis (see Table 3).

5.4.2 Impact of the noradrenergic system and the HPA axis on trauma film specific memory

Salivary cortisol levels and sAA measured in both original studies proved the efficacy of the pharmacological treatments on noradrenergic activation/inhibition and HPA axis activation (Rombold et al., 2016a; Rombold et al., 2016b). Concerning forced choice recognition, a significant effect of film sequence ($F_{df2,336}$ = 122.61, p < .01, η_p^2 = .42) was revealed, while there was no effect of intervention ($F_{df3,168}$ = 1.36, p = .26, η_p^2 = .02). The film sequence x intervention interaction effect (see Figure 15) was significant ($F_{df6,336}$ = 2.21, p = .04, η_p^2 = .04), indicating a differential influence of intervention on the number of correct pre-, peri-, and post-trauma film scene memories. Post-hoc tests revealed that clonidine led to significantly less correct peri-trauma film memories compared to placebo (p = .01) and that clonidine led to less correct peri-trauma film memories compared to yohimbine on a trend level (p = .08). The hydrocortisone group did not differ significantly from any of the other groups.

Including the DSS-Acute (Stiglmayr et al., 2003) as covariate in the general linear model did not modify the significance of the interaction effect (film sequence x intervention).

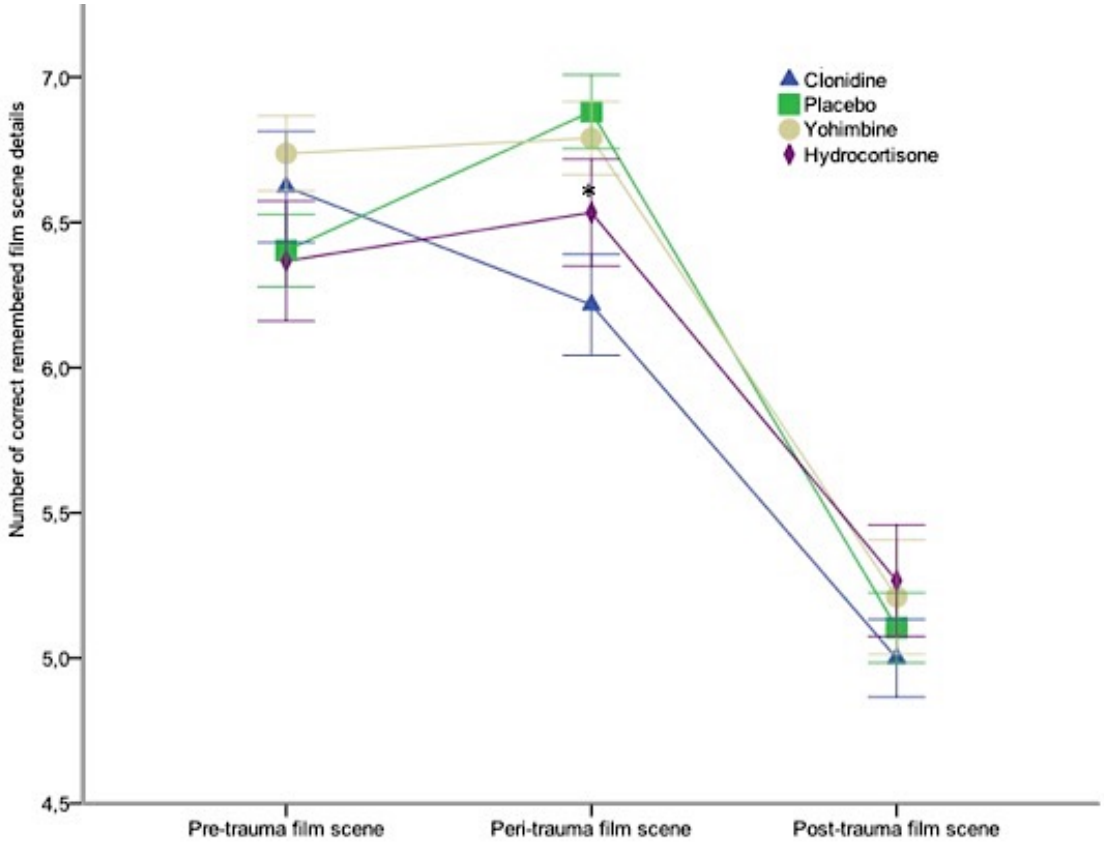

Figure 13. Number of correct remembered pre-, peri-, and post-trauma film scene details assessed after seven days for the clonidine, yohimbine, hydrocortisone, and placebo group. Significant difference between the clonidine and placebo group (* $p < .05$). Error bars represent +/- 1 SE.

5.5 Conclusions

The current study examined the impact of the noradrenergic system and the HPA axis on forced choice recognition after a distressing film in healthy women. A differential influence of intervention on the number of correct pre-, peri-, and post-trauma film scene memories after one week was found. We revealed a selective effect for recognition of peri-trauma film scene details compared to pre- and post-trauma film scene details. Clonidine led to significantly fewer correct peri-trauma film scene memories compared to placebo. Further, clonidine led to fewer correct peri-trauma film scene memories compared to yohimbine on a trend level.

The results of this study indicate that pharmacologically inhibited noradrenergic activity during a distressing film impacts subsequent recognition of

film details. The results concur with data on fear expression in rodents where clonidine administration has led to weakened memory consolidation compared to yohimbine (Gazarini, Stern, Carobrez, & Bertoglio, 2013a) and with data on impaired emotional memory formation after beta-blocker administration in healthy participants (Chamberlain & Robbins, 2013). For example, one of the first studies assessing the impact of blocking the β-adrenergic receptor with the beta-blocker propranolol found impaired memory of a slides series accompanied by an emotional narrative compared to placebo (Cahill et al., 1994). Further, participants who received either placebo or propranolol before learning 38 word lists of 24 nouns including one emotional noun showed enhanced memory for emotional nouns compared to neutral nouns and impaired memory for neutral nouns directly preceding emotional nouns after placebo administration (Strange et al., 2003). After propranolol administration, memory for emotional nouns did not differ from memory for neutral nouns and neutral nouns directly preceding emotional nouns were remembered better than neutral control nouns (Strange et al., 2003).

The negative findings on pharmacologically enhanced noradrenergic activity and HPA axis activation are not in line with previous data which have shown that noradrenergic activation and HPA axis activation lead to increased emotional memory formation (Roozendaal & McGaugh, 2011; Schwabe et al., 2012). The effect might be weakened by assessing forced choice recognition in healthy participants and not in PTSD patients. For example, in PTSD patients disorganization in trauma memories is more increased than in memories of unpleasant events compared to trauma survivors without PTSD (Jelinek *et al*, 2009). Further, the forced choice recognition paradigm instead of intentional free recall could be responsible.

In models of PTSD, the relationship between the perceptual memory system leading to intrusive memories and the episodic memory system responsible for recognition is disputed (Brewin, 2014). Some theories suggest that intrusive memories of traumatic events are unrelated or even inversely related to voluntary memory of trauma details (Jacobs & Nadel, 1985; Metcalfe & Jacobs, 1998). Others suggest a positive correlation between involuntary memory of the trauma and recognition of trauma details (Ferree & Cahill, 2009). The dual representation theory of PTSD (Brewin *et al*, 1996; Brewin *et al*, 2010) postulates two different,

parallel memory mechanisms. Perceptual memories of sensational representations (S-reps) are activated automatically by specific cues. In parallel, there are contextualized representations (C-reps) that are selective and in line with conscious attention. They can be retrieved voluntarily or automatically. S-reps support intrusive memories, whereas C-reps support episodic memories and intentional recall of traumatic memory. Intentional recall deficits of trauma-details six months (Halligan et al., 2003) and one year (Rothbaum et al., 1992) after the event have been associated with PTSD diagnosis in assault victims. PTSD patients show greater disorganization in trauma memories compared to unpleasant but non-traumatic memories (Brewin *et al*, 2014). In the current study, clonidine led to significantly fewer correct peri-trauma film scene memories compared to placebo. However, no difference in recognition between yohimbine and placebo was found. In a previous study, no difference in intrusive memories between the clonidine and placebo group was found and after yohimbine administration the number of intrusive memories showed a delayed decrease compared to clonidine and placebo over the four days following the trauma film (Rombold *et al*, 2016a). Therefore, the noradrenergic system seems to act differently on recognition (C-reps) and intrusive memories (S-reps) after a distressing film. Noradrenergic activation led to a delayed decrease of intrusive memories, whereas noradrenergic inhibition led to impaired recognition. Our data indicate that neither noradrenergic activation nor noradrenergic inhibition results in an invers relationship of intrusive memories and voluntary memories. Our findings are more in line with emotional memory formation (Gazarini et al., 2013a) than with the dual representation theory of PTSD (Brewin *et al*, 2014). However, the fact that our healthy participants did not suffer from PTSD after the distressing film might be responsible for the findings on the relationship between intrusive memories and recognition.

The study shows major strengths: assessing the influence of noradrenergic activation, noradrenergic inhibition, and HPA axis activation *during* a distressing film on consecutive emotional memory recognition; examining pre-, peri-, and post-trauma film scene memory separately; using a placebo-controlled design. At the same time, several limitations should be considered. The influence of stress during real trauma on emotional memory recognition could differ from its

influence during a distressing film. Further, it is not known whether the findings are only applicable to young, healthy women or also to trauma survivors who develop PTSD. As we only included women, the results might not be transferable to men (Felmingham et al., 2012; Strange et al., 2003). Further, the null finding in the yohimbine and hydrocortisone groups may be due to ceiling effects. Finally, the pre-, peri-, and post-trauma film scenes are clearly distinguishable in terms of encoding. Consolidation processes, however, might overlap.

In summary, noradrenergic inhibition (clonidine) led to significantly fewer correct peri-trauma film scene memories compared to placebo and to fewer correct peri-trauma film scene memories compared to noradrenergic activation (yohimbine) on a trend level. Compared to placebo, no differences in recognition of peri-trauma film details after noradrenergic activation with yohimbine and HPA axis activation with hydrocortisone were found. The results support part of our hypotheses and show that noradrenergic inhibition during trauma film leads to reduced emotional memory recognition for peri-trauma film details.

6 Lower Heart Rate Variability at Baseline is Associated With More Consecutive Intrusive Memories in an Experimental Distressing Film Paradigm (Study IV)

This chapter has been published as 'Rombold-Bruehl, F., Otte, C., Renneberg, B., Schmied, A., Zimmermann-Viehoff, F., Wingenfeld, K., & Roepke, S. (2017). Lower heart rate variability at baseline is associated with more consecutive intrusive memories in an experimental distressing film paradigm. *The World Journal of Biological Psychiatry*, 1-6.'

DOI: http://dx.doi.org/10.1080/15622975.2017.1372628

7 General Discussion

In the following chapter the findings, gain of knowledge, and conclusions of this dissertation project on neuropsychoendocrinological and ANS influences on intrusion formation and memory for trauma film details will be discussed.

In section 7.1 the main results from the four studies will be summarized and discussed. Next, in section 7.2 the main strengths and limitations of the four studies will be summarized and discussed. Further, in section 7.3 implications for models of PTSD that result from the findings across all studies will be outlined and included in existing models of PTSD. In section 7.4 clinical implications derived from the results across all studies will be made. Additionally, in section 7.5 further suggestions for future research on trauma related memories will be made. Finally, section 7.6 will end this chapter with general conclusions on intrusive memory formation and voluntary trauma memory.

7.1 Discussion of the main findings

7.1.1 The stress response systems and intrusive memories

In study I during encoding and consolidation of the trauma film yohimbine compared to clonidine and placebo led to a delayed decrease in the number and vividness of intrusions over the four days following the trauma film. Although, study I assessed the influence of the noradrenergic system on consecutive intrusive memories by manipulating the activity of the noradrenergic system during encoding and consolidation for the first time, the influence of stress on emotional memory formation has been in the focus of psychoneuroendocrinology research for longer. The presented findings are in line with data from rodent studies that show an influence of increased noradrenergic activity during consolidation on enhanced fear memory (Dębiec et al., 2011; Diaz-Mataix et al., 2017; Gazarini et al., 2013b). Additionally, the findings are in line with the influence of stress during consolidation on emotional memory formation in

healthy participants (Segal & Cahill, 2009; Smeets et al., 2008). In study I salivary cortisol levels increased 15 minutes after yohimbine administration[8]. This might have had an impact on the findings as catecholamine-glucocorticoid interactions seem to be crucial for emotional memory formation (Roozendaal & McGaugh, 2011; Segal et al., 2014). The results extend findings from a study which assessed the influence of stress during consolidation of negative images on unintentional memories of these images (Bryant et al., 2013). An interaction of increased sAA activity and increased salivary cortisol levels was found to be associated with an increased number of subsequent unintentional memories of the images (Bryant et al., 2013). However, this was only the case for men and not for women (Bryant et al., 2013). Further, a previous trauma film paradigm study assessed the influence of endogenous sAA and salivary cortisol during encoding and consolidation of a trauma film and found no influence of sAA activity on number and vividness of intrusions (Chou et al., 2014). However, post-trauma film salivary cortisol levels were associated with the number of intrusive memories in participants with enhanced sAA activity (Chou et al., 2014).

The clonidine and placebo group did not differ in number or vividness of intrusions. This finding indicates no influence of noradrenergic inhibition during trauma on subsequent intrusive memories. This finding contradicts data on reduced memory performance after β-adrenergic blockade in humans for emotional and non-emotional material (Kuffel et al., 2014; Strange et al., 2003; van Stegeren, 2008). The lacking effect of clonidine contradicts the hypotheses and could be explained by a floor effect of the intensity of the stressor, a relatively mild stressor compared to real trauma, or by a missing difference between HPA axis activation in the clonidine and placebo group. Further, the experienced degree of distress caused by the intrusions did not differ between the yohimbine, clonidine, and placebo groups which indicates that there was no effect of the noradrengic system on the degree of distress caused by the intrusions.

[8] The influence of yohimbine administration in healthy subjects on salivary cortisol levels has so far been contradictory as enhancement of salivary cortisol levels has been observed (Sommer et al., 2011) as well as no effect of yohimbine administration on salivary cortisol levels (Gurguis et al., 1997).

In study II during encoding and consolidation of the trauma film hydrocortisone compared to placebo did not affect the number and vividness of intrusions nor the distress caused by the intrusions. The results show that the biological mechanisms for intrusion formation seem to be different from glucocorticoid regulated emotional memory formation in healthy humans and rodents (de Quervain, Schwabe, & Roozendaal, 2017; McGaugh & Roozendaal, 2002). For example, the administration of glucocorticoids or synthetic GR ligands, like the substance dexamethasone before or after training/encoding, enhances emotional memory consolidation in rats and humans (de Quervain et al., 2017). Further, cortisol levels during encoding and consolidation of emotional material are positively related to subsequent memory performance on that material (Roozendaal & McGaugh, 2011; Wingenfeld & Wolf, 2015; Wolf, 2009). In addition, following hydrocortisone administration increased cortisol levels during consolidation led to better emotional memory in healthy participants (Buchanan & Lovallo, 2001; Kuhlmann & Wolf, 2006). Of note, the administration of moderate doses of glucocorticoids enhances memory, whereas low and high doses are mostly not as effective and even impair memory consolidation (Andreano & Cahill, 2006; Roozendaal, Williams, & McGaugh, 1999). In contrast, the administration of metyrapone to block cortisol synthesis during TSST and a following picture story impaired memory consolidation of that picture story in comparison to placebo and beta-blocker propranolol (Maheu et al., 2004).

Enhanced glucocorticoid driven emotional memory consolidation was recently shown to depend on arousal-induced noradrenergic activation in the amygdala and its interactions with other brain regions, such as the hippocampus and the cortex (de Quervain et al., 2017; Roozendaal & McGaugh, 2011). This synergistic effect of the HPA axis and the noradrenergic system is supported by findings in rodents, in which β-adrenoceptor antagonists, which were either systemically administered or administered directly into the basolateral amygdala, blocked glucocorticoid driven emotional memory enhancement effects (Quirarte, Roozendaal, & McGaugh, 1997; Roozendaal, Okuda, Van der Zee, & McGaugh, 2006b). In humans, a recent finding in healthy women suggests emotional memory enhancement after hydrocortisone administration only when endogenous sAA was additionally increased during consolidation (Segal et al., 2014). However, in study

II, even after including endogenous sAA activity in the analysis, hydrocortisone administration did still not influence intrusion formation. A reason for the missing synergistic effect of the HPA axis and the noradrenergic system in study II could either be a different underlying mechanism of intrusion formation compared to emotional memory or an insufficient elevation of noradrenergic arousal. Taking the results of study I and recent developments on emotional memory (de Quervain et al., 2017) into account, it seems more likely that intrusion formation depends on similar mechanisms as emotional memory formation and that sAA activity was not high enough in the hydrocortisone group to impact on intrusions.

Findings on endogenous cortisol during consolidation and subsequent intrusive memories have been contradictory (Bryant et al., 2013; Chou et al., 2014; Nicholson et al., 2014). In line with findings on emotional memory consolidation (Segal et al., 2014) a trauma film paradigm study showed that post-trauma film endogenous salivary cortisol levels and the number of consecutive intrusions were associated in participants which showed enhanced endogenous sAA activity during the trauma film (Chou et al., 2014). However, gender differences should be taken into account since the synergistic effect of increased noradrenergic activity and increased cortisol levels on retrospectively assessed intrusion formation was found only in men (Bryant et al., 2013). In line with the results from study II, in healthy female and male participants no association was found between endogenous cortisol and retrospectively assessed intrusion formation of negatively valenced pictures (Nicholson et al., 2014). However, in PTSD patients with enhanced endogenous sAA activity during consolidation of negatively valenced pictures endogenous cortisol was positively associated with retrospectively assessed intrusion formation (Nicholson et al., 2014).

The lacking effect of cortisol could also depend on the time it was administered since recent research indicates a biphasic time-dependent effect of glucocorticoids on emotional memory formation (de Quervain et al., 2017). Shortly after cortisol administration fast, non-genomic effects take place which are followed by the slower, genomic effects starting around 60 min later (Cornelisse et al., 2014). Therefore, the results of study II do not allow the conclusion that cortisol does not influence intrusion formation. It can only be concluded that hydrocortisone administration 60 min before a stressor does not influence

subsequent intrusions. Genomic cortisol effects might still impact on intrusion formation.

Furthermore, genetic and epigenetic influences on intrusive memory formation might impact on intrusion formation (Cheung & Bryant, 2015). The HSP90 co-chaperone FK506 binding protein five (FKBP5) influences cortisol response to stress (Mahon, Zandi, Potash, Nestadt, & Wand, 2013). Differences in FKBP5 genetic polymorphisms have been shown to influence intrusion formation of negatively valenced and neutral pictures in healthy participants (Cheung & Bryant, 2015). Participants defined as high-risk allele carriers after two days retrospectively reported more intrusive memories than participants defined as low-risk allele carriers (Cheung & Bryant, 2015). However, intrusions and increase in cortisol were not associated (Cheung & Bryant, 2015) which would again be in line with the results of the here presented study. Therefore, other mechanisms associated with FKBP5 genetic polymorphisms could be involved in intrusion formation.

Preliminary case study findings indicated a treatment effect of cortisol on intrusive memories (Aerni et al., 2004). However, concluding from two recent studies in healthy participants (Graebener, Michael, Holz, & Lass-Hennemann, 2017) and PTSD patients (Ludäscher et al., 2015) cortisol does not seem to effect intrusive memories. In a recent trauma film paradigm study the impact of cortisol administration on the three days following the trauma film was assessed within a randomized, double-blind, placebo controlled design and no effect of hydrocortisone on the number of intrusions was found (Graebener et al., 2017). Further, in a recent randomized, double-blind, placebo-controlled, crossover design treatment study in female PTSD patients neither 10 mg nor 30 mg hydrocortisone, each administered for one week, showed an effect on the frequency and intensity of intrusive memories (Ludäscher et al., 2015). However, the generalizability of the findings is limited since the sample was chronically traumatized, psychotropically medicated, and showed a high number of comorbid disorders (Ludäscher et al., 2015).

The findings indicate that there is no influence of hydrocortisone administered 60 minutes prior to the trauma film on subsequent intrusion formation. However, noradrenergic activation in contrast to noradrenergic

inhibition during trauma seems to influence subsequent intrusion formation as indicated by a delayed decrease of intrusions.

7.1.2 The stress response systems and memory of trauma film details

In study III during encoding and consolidation of the trauma film a differential influence of intervention (clonidine vs. yohimbine vs. hydrocortisone vs. placebo) on the number of correctly remembered pre-, peri-, and post-trauma film scene details was found. Clonidine administration resulted in significantly fewer correct peri-trauma film scene memories compared to placebo and in fewer correct peri-trauma film scene memories compared to yohimbine on a trend level. There was no difference between the hydrocortisone, the yohimbine, and the placebo group.

The results of study III show an impairing effect of clonidine on memories of peri-trauma film scenes. This result concurs with findings in rodents which show that clonidine compared to yohimbine weakens fear memory consolidation (Gazarini et al., 2013b). Further, the results are in line with findings in healthy humans which show impaired memory formation for emotional material after blocking the beta-adrenergic receptor (Chamberlain & Robbins, 2013). For example, Cahill and colleagues (1994) found memory impairments for presented slides and matching emotional narratives after the administration of beta-blocker compared to placebo. However, the lacking effect of yohimbine and hydrocortisone is not in line with findings which show that increased noradrenergic activity and HPA axis activation enhance memory formation for emotional material in humans (Roozendaal & McGaugh, 2011; Schwabe et al., 2012). This might be due to ceiling effects since the healthy participants in general performed well in the forced choice recognition test.

The results suggest that noradrenergic inhibition during trauma might impair subsequent memory for peri-traumatic details. No effect of noradrenergic activation and HPA axis activation during the trauma film on memory for trauma film details was found in this paradigm.

7.1.3 Heart rate variability and intrusive memories

In study IV lower baseline HRV (indicated by higher LF/HF ratio) before the trauma film was significantly associated with more intrusions. Lower baseline HRV (HF) before the trauma film was associated with more intrusions on trend level. Further, significant time x HRV (HF and LF/HF ratio) interactions were found indicating a different time course of intrusive memories after the trauma film depending on baseline HF and baseline LF/HF ratio components of HRV.

Studies on the impact of HRV on intrusive memories or PTSD are limited. The results of study IV extend recent findings on intrusion formation as cognitive control deficits, a risk factor for intrusion formation, are mediated by HRV (Gillie et al., 2015). Lower HRV seems to be associated with less self-regulation abilities (Geisler et al., 2013) and with less cognitive function (Gillie et al., 2014, 2015; Thayer et al., 2009). Further, the results of study IV extend the findings of two military cohort studies that reported an association between lower pre-deployment HRV and post-deployment PTSD symptoms (Minassian et al., 2015; Pyne et al., 2016). Mineassian and colleagues (2015) found an association between reduced HRV (indicated by higher LF/HF ratio) before combat deployment and an increased PTSD risk following deployment in male marines. Accordingly, Pyne and colleagues (2016) found that lower HRV (HF) before deployment predicts PTSD symptoms after deployment. However, this was only the case for soldiers who already showed higher PTSD symptoms before deployment. In study IV an association between lower baseline HRV and an increased number of consecutive intrusive memories was found in healthy young women, just as Minassian and colleagues (2015) and Pyne and colleagues (Pyne et al., 2016) found low pre-deployment HRV in male marines to be associated with post-deployment PTSD. Therefore, evidence is increasing that lower HRV before a trauma is associated with higher risk for PTSD and intrusive memories.

The findings indicate that lower baseline HRV maybe a vulnerability factor for consecutive intrusions after trauma and that women with lower baseline HRV recover slower from trauma, as indicated by a delayed decrease in intrusive memories.

7.2 General strengths and limitations of the studies

The major strength of the studies lies in the randomized controlled design which enables the induction of intrusive memories and the assessment of memory recognition. Compared to traumatic real life events the trauma film used was relatively mild and therefore it is uncertain if the results are transferrable to PTSD patients. However, according to DSM-5 the definition of trauma has been extended and now includes indirect exposure to trauma details in a work context via electronic media and television (American Psychiatric Association, 2013). This argues for the use of trauma film paradigms and the comparability of the intrusive memories caused and those following real life trauma. The studies are limited because the results are not transferrable to men since the samples were restricted to women and sex hormones are assumed to influence intrusion formation (Cheung & Bryant, 2015; Ferree et al., 2011). Further, intrusive memories were assessed via a self-report diary and might, therefore, be inaccurate due to difficulties in detecting the intrusions and participants not reporting them (Takarangi et al., 2014). However, the additional online diary and daily reminder via text message increased participants' compliance. For further discussion of the limitations please see sections 3.4, 4.5, 5.5, and 6.5.

7.3 Implications for models of intrusive memories

Up to now, the most elaborate PTSD models focus on cognitive processes such as dissociation during trauma, fragmentation of trauma memory, and poor contextualization into autobiographical memory (Brewin et al., 1996; Brewin et al., 2010; Ehlers & Clark, 2000; Ehlers et al., 2004). However, the revised dual representation theory (Brewin et al., 2010) and the overconsolidation model of PTSD (Pitman & Delahunty, 2005) include neurobiological processes. The studies presented in this dissertation strongly advocate the inclusion of peri- and post-trauma neurobiological and pre-trauma physiological parameters in the models of PTSD. In the following, I will embed the knowledge gained from studies I, II , III, and IV into the theories of PTSD and thereby extend these models.

7.3.1 Cognitive models of PTSD

The cognitive model of PTSD (Ehlers & Clark, 2000) suggests that patients with PTSD experience a sense of current threat, show excessively negative appraisals of the traumatic event, and show disturbances in autobiographical memory. The latter includes three general memory processes as a source for the formation of intrusive memories. These are increased perceptual priming, decreased memory elaboration, and increased associative learning. The model hypothesizes that arousal during trauma encoding results in fragmentation of the memories integrated into the autobiographical memory system, as reflected by the occurrence of intrusive memories. Ehlers and Clark (2000) differentiate between conceptual and data-driven processing. Trauma victims who report clear thinking and analyzing thoughts during trauma are assumed to encode the situation conceptually, whereas trauma victims who report confusion and strong sensory impressions during trauma are assumed to encode the situation in a data-driven way (Ehlers & Clark, 2000). However, the model does not explain the underlying neurobiological mechanisms.

The dual representation theory (Brewin et al., 2010) postulates the existence of verbally accessible memory (VAM system) and situationally accessible memory (SAM system). The VAM system includes abstract declarative representations enbeded in the autobiographical context. This information can be retrieved deliberately. The SAM system includes sensory, affection, and emotion bound representations. This information is triggered involuntarily by cues. In PTSD the traumatic event is stored in the SAM system and the representation in the VAM system is impaired. The SAM system is supposed to be controlled by the amygdala, whereas the VAM system is controlled by the hippocampus. The amygdala controlled processes (e.g., emotional appraisal and recognition of situations and appraisal of potential threat) are assumed to inhibit processes controlled by the hippocampus (e.g., memory consolidation).

The cognitive model of PTSD and the dual representation theory should be extended by neuroenocrinological mechanisms. As indicated by the results of this dissertation a synergistic effect of increased noradrenergic activity and increased cortisol levels during trauma might contribute to a disturbed integration of the

trauma memory trace into autobiographical memory. An increased release of stress hormones during trauma might support the data-driven processing resulting in increased perceptual priming, decreased memory elaboration, and increased associative learning. In the following a hypothetical neuroendocrine model is introduced: traumatic events cause stress and lead to a fast activation of the sympathetic nervous system. The impact of noradrenaline in the basolateral complex of the amygdala is enhanced. The amygdala stores emotional information and interacts with different brain regions responsible for learning and memory such as the prefrontal cortex and the hippocampus (McGaugh, 2013). In particular, the amygdala is responsible for processing sensory information (Jovanovic & Ressler, 2010) and the lateral nucleus of the amygdala serves as its sensory gateway by receiving visual, auditory, gustatory, somatosensory, and olfactory information (Rodrigues, LeDoux, & Sapolsky, 2009). Increased activity of the noradrenergic system during trauma could be responsible for stronger S-reps and impaired C-reps resulting in a later reduction of intrusive memories. Trauma survivors with increased noradrenergic activity during trauma might be at higher risk for sensory-bound information processing which supports the occurence of subsequent involuntary intrusive memories.

Noradrenergic inhibition during encoding and consolidation of the trauma film led to significantly less correct peri-trauma film scene memories compared to placebo. Further, noradrenergic inhibition led to significantly less correct peri-trauma film scene memories compared to noradrenergic activation on a trend level (study III). No difference between noradrenergic activation, HPA axis activation, and placebo was found. In contrast, in study I noradrenergic activation led to a delayed decrease of intrusions compared to clonidine and placebo while no difference between the number of intrusions following noradrenergic inhibition and placebo was found. Ehlers & Clark (2000) propose the coexistence of intrusive memories and impaired voluntary retrieval of trauma details and functionally independent perceptual and episodic memory systems. Data from trauma film paradigm studies using a cognitive task underscore the assumption of different mechanisms resulting in involuntary and voluntary memory of the trauma film (James et al., 2016). For example, the number of intrusions is reduced by visuospatial tasks while recognition memory is not impacted (Holmes et al., 2010).

Intentional recall deficits caused by memory fragmentation (Foa & Rothbaum, 2001), impaired memory representation (Brewin et al., 1996), or strong perceptual priming (Ehlers & Clark, 2000) are proposed by several theories while others propose that voluntary and involuntary memories are enhanced (Berntsen, 2011; Rubin, Boals, & Berntsen, 2008). According to Ehlers (2010) light is shed on the controversy about the relationship of voluntary and involuntary memory of the trauma by looking at the trauma in a more differentiated way and separating it into a sequence of events instead of defining it as one event (Ehlers, 2010).

As a psychophysiological parameter, lower baseline HRV was found to be associated with more subsequent intrusive memories (study IV). Lower HF, an indicator for less parasympathetic activity, before a trauma seems to be a vulnerability factor for consecutive intrusions. This is in line with the findings from study I that indicated a link between increased sAA activity, an indicator of sympathetic activation, during encoding and consolidation and a delayed decrease of the number and vividness of intrusions. The sympathetic nervous system and the parasympathetic nervous system are both part of the autonomic nervous system and operate in a complementary way. The findings from studies I and IV seem to be associated as they both suggest a link between predominat sympathetic nervous system activity before (study IV) and during (study I) trauma and subsequent intrusive memories. As indicated by two recent studies (Minassian et al., 2015; Pyne et al., 2016) and the findings of study IV, baseline HRV seems to be a pre-existing vulnerability factor for intrusive memories and PTSD that should be taken into consideration for the etiological models of PTSD. The implications of this dissertation project for models of PTSD are displayed in Figure 16.

7.3.2 Overconsolidation model of PTSD (Pitman & Delahunty, 2005)

The memory overconsolidation model of PTSD by Pitman and Delahunty (2005) postulates that intrusions are the result of memory overconsolidation. In this model, the release of noradrenaline and cortisol (unconditioned response) during trauma (unconditioned stimulus) results in an overconsolidation of the traumatic memories and in a poorer contextualization of trauma memories. Memory traces are strengthened and primed more easily, as reflected in an increased number of

intrusions. After the traumatic event, trauma reminders (conditioned stimulus) release stress hormones (conditioned response) which cause even more overconsolidation. In study I noradrenergic activation led to a delayed decrease in the number and vividness of intrusions compared to noradrenergic inhibition and placebo. However, noradrenergic activation via yohimbine led to an increase of cortisol levels. Therefore, the increase in intrusions and the vividness of intrusions could not solely be associated with the noradrenergic system. This finding is in line with the results from a previous trauma film paradigm study which found a positive correlation of intrusions and increased salivary cortisol levels only in those participants with increased sAA activity (Chou et al., 2014).

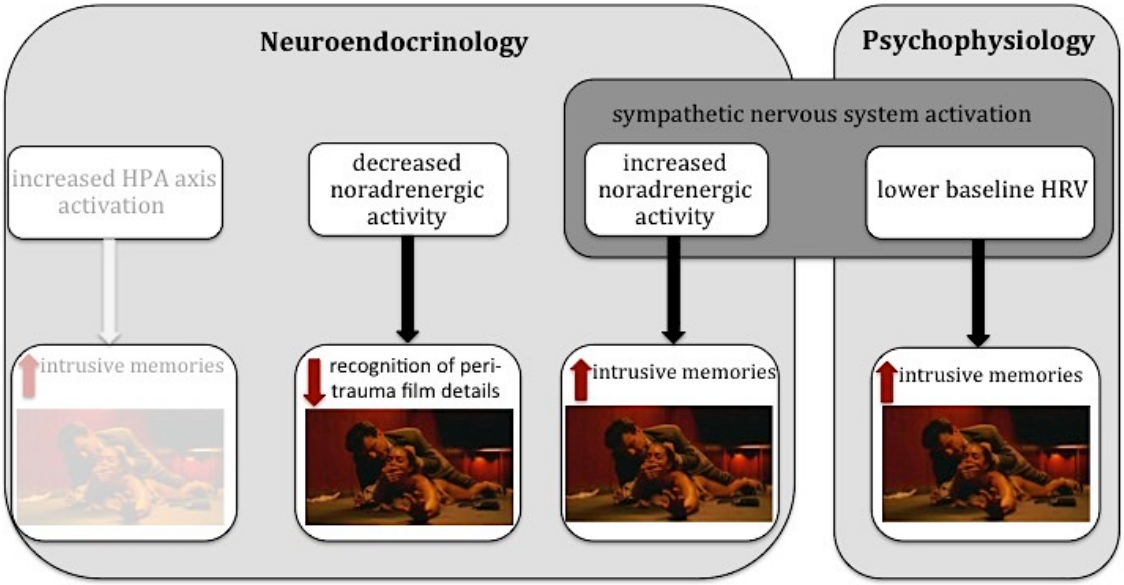

Figure 16. Summary of the main findings on trauma related memories.

7.4 Clinical implications

Basic research findings on stress and memory have successfully been translated into clinical interventions. For example, the administration of glucocorticoids after trauma has, so far, turned out to be the only effective pharmacological intervention to prevent PTSD (Amos, Stein, & Ipser, 2014; Sijbrandij et al., 2015). Clinical implications from the studies of this dissertation project will be outlined in the following section.

First, individuals who show an increased activity of the noradrenergic system during trauma and its consolidation phase might recover from intrusive memories at a slower rate. The occurence of intrusive memories during the initial time after trauma predicts subsequent PTSD diagnosis (Ehlers, 2010; O'Donnell, Elliott, Lau, & Creamer, 2007; Shalev et al., 1996). Subsequently, trauma survivors with an increased activity of the noradrenergic system during trauma and a delayed decrease of intrusive memories might be at higher risk for subsequent PTSD.

Second, individuals with decreased noradrenergic activity during trauma and its consolidation phase might recognize less trauma related details. This finding is important for perpetrator confrontations and exposure therapy for PTSD.

Third, individuals with lower HRV before a traumatic event seem to be more vulnerable for intrusive memories following trauma and might therefore be at higher risk for PTSD. The significant association of lower baseline HRV and more subsequent intrusive memories could serve as a target for the development of prevention mechanisms. Pre-trauma assessments in high-risk professional groups (soldiers, firemen, first aid workers) could identify those with lower HRV and therefore increased risk for the development of intrusive memories. Those at risk could receive HRV biofeedback training to increase their HRV and decrease the potential risk for intrusive memories and PTSD. First evidence indicates successful relaxation training for veterans with PTSD which enabled them to voluntarily increase their own HRV (Lewis et al., 2015). This resulted in improved attention and short-term memory (Ginsberg, Berry, & Powell, 2010).

Fourth, severe medical illnesses such as traumatic brain injury or acute respiratory distress syndrome followed by intensive care unit treatment can result in the development of subsequent PTSD with incident rates comparable to other trauma exposure (Kapfhammer, Rothenhausler, Krauseneck, Stoll, & Schelling, 2004; Krauseneck, Rothenhausler, Schelling, & Kapfhammer, 2005; Stoll et al., 2000; Tedstone & Tarrier, 2003). The major risk factor for the development of PTSD in these patients are intensive care unit treatment-related traumatic memories (Kapfhammer et al., 2004; Schelling et al., 2003). Cortisol administration is common during intensive care unit treatment. The results of study II do not

imply an association of increased risk for intrusive memories following cortisol administration in intensive care unit patients.

Overall, the findings contribute to a growth of clinically relevant knowledge on trauma related memory and PTSD. The conducted trauma film paradigm studies provided the opportunity of studying mechanisms that could not have been studied in real life trauma and consequentially drawing clinical implications.

7.5 Future research

Future studies should continue the search for neurobiological discriminative factors of those who develop more intrusive memories after trauma and those who develop less intrusive memories and are therefore at lower risk for PTSD.

It would be desirable to systematically assess the influence of the HPA axis and the noradrenergic system on different aspects of voluntary recall, such as free recall and forced choice recognition paradigms, to examine whether they are influenced in a different way. The distinction of different aspects of voluntary memory could shed light on the versatile voluntary memory impairments in PTSD patients. Memory measures within trauma film paradigms have received little attention (James et al., 2016). Questionnaires with good validity and reliability to measure different voluntary memory aspects, such as recognition and free recall of trauma film details, should be designed.

Further, the influence of noradrenergic-, glucocorticoid-, and endocannabinoid signaling on intrusion formation has to be better understood. The endocannabinoid system predominantly regulates the release of the neurotransmitters gamma-aminobutyric acid (GABA) and glutamate (Freund, Katona, & Piomelli, 2003). Recently, the endocannabinoid system has been shown to mediate glucocorticoid driven memory consolidation (Balsevich, Petrie, & Hill, 2017; de Quervain et al., 2017). For example, enhanced memory consolidation following glucocorticoid administration is prevented by the blockade of cannabinoid type one receptors in the amygdala and the hippocampus (Atsak et al., 2015; Campolongo et al., 2009; de Oliveira Alvares et al., 2010). Therefore, a potential impact of the endocannabinoid system on intrusive memories needs to be assessed.

Further, genetic and epigenetic influences on intrusive memory formation should be considered in future studies as they might impact on intrusion formation (Cheung & Bryant, 2015). For example, increased cortisol has been associated with one nucleotide polymorphism of the glucocorticoid receptor gene (van Rossum et al., 2006) which has approximately 14 % West-European carriers (van Rossum et al., 2003). Carriers of this nucleotide polymorphism who underwent cardiac surgery developed more PTSD symptoms one week after surgery and more traumatic memories six months later compared to non-carriers (Hauer et al., 2011). Further, genetic differences have also been associated with emotional memory in healthy subjects (Ackermann, Heck, Rasch, Papassotiropoulos, & de Quervain, 2013).

Finally, the difference of pharmacologically induced stress through hydrocortisone and yohimbine administration and more naturally elevated stress on intrusion formation is of interest. Our research group is currently assessing the influence of the Trier Social Stress Test (TSST) on the formation of intrusive memories. After a preparation phase, participants perform a speech in front of an audience and an arithmetic task. The HPA axis is reliably activated by the TSST (Kirschbaum, Pirke, & Hellhammer, 1993).

7.6 Conclusions

To summarize, the findings of this dissertation project suggest an influence of noradrenergic activation on intrusion formation, as indicated by a delayed decrease of intrusions. Noradrenergic inhibition and HPA axis activation, however, do not seem to influence consecutive intrusion formation. Further, the results suggest an impairing effect of noradrenergic inhibition on recognition of trauma details. Finally, the findings provide support for an association between lower HRV before a traumatic event and an increased number of intrusions after trauma. In summary, the predominat activation of the sympathetic nervous system before or during trauma seems to be associated with subsequent intrusive memories. The results of this dissertation project shed light on neuropsychoendocrinological and psychophysiological aspects of intrusive memories and voluntary memory of trauma details.

Alle Erinnerung ist Gegenwart.

Novalis (1772 – 1801)

8 References

Ackermann, S., Heck, A., Rasch, B., Papassotiropoulos, A., & de Quervain, D. J. (2013). The BclI polymorphism of the glucocorticoid receptor gene is associated with emotional memory performance in healthy individuals. *Psychoneuroendocrinology, 38*(7), 1203-1207.

Aerni, A., Traber, R., Hock, C., Roozendaal, B., Schelling, G., Papassotiropoulos, A., . . . Dominique, J. (2004). Low-dose cortisol for symptoms of posttraumatic stress disorder. *American Journal of Psychiatry, 161*(8), 1488-1490.

American Psychiatric Association. (2013). *Diagnostic and Statistical Manual of Mental Disorders (DSM-5®)*. Washington, DC: American Psychiatric Association.

Amos, T., Stein, D. J., & Ipser, J. C. (2014). Pharmacological interventions for preventing post-traumatic stress disorder (PTSD). *Cochrane Database of Systematic Reviews, 7*, 1-66.

Andreano, J. M., & Cahill, L. (2006). Glucocorticoid release and memory consolidation in men and women. *Psychological Science, 17*(6), 466-470.

Andreano, J. M., & Cahill, L. (2009). Sex influences on the neurobiology of learning and memory. *Learning and Memory, 16*(4), 248-266.

Arntz, A., de Groot, C., & Kindt, M. (2005). Emotional memory is perceptual. *Journal of Behavior Therapy and Experimental Psychiatry, 36*(1), 19-34.

Atsak, P., Hauer, D., Campolongo, P., Schelling, G., Fornari, R. V., & Roozendaal, B. (2015). Endocannabinoid signaling within the basolateral amygdala integrates multiple stress hormone effects on memory consolidation. *Neuropsychopharmacology, 40*(6), 1485-1494.

Bailey, C. R., Cordell, E., Sobin, S. M., & Neumeister, A. (2013). Recent progress in understanding the pathophysiology of post-traumatic stress disorder. *CNS Drugs, 27*(3), 221-232.

Balsevich, G., Petrie, G. N., & Hill, M. N. (2017). Endocannabinoids: Effectors of glucocorticoid signaling. *Frontiers in Neuroendocrinology, 47*, 86-108.

Beauchaine, T. P., & Thayer, J. F. (2015). Heart rate variability as a transdiagnostic biomarker of psychopathology. *International Journal of Psychophysiology, 98*(2), 338-350.

Beck, A. T. (1978). The depression inventory. *Philadelphia: Center for Cognitive Therapy.*

Bedard-Gilligan, M., & Zoellner, L. A. (2012). Dissociation and memory fragmentation in post-traumatic stress disorder: an evaluation of the dissociative encoding hypothesis. *Memory, 20*(3), 277-299.

Bernstein, D. P., & Fink, L. (1998). *Childhood trauma questionnaire: A retrospective self-report: Manual.* San Antonio, TX: Psychological Corporation.

Berntsen, D. (2011). Involuntary autobiographical memories. *Applied Cognitive Psychology, 10*(5), 435-454.

Billman, G. E. (2013). The LF/HF ratio does not accurately measure cardiac sympatho-vagal balance. *Frontiers in Physiology, 4*, 26.

Bravi, A., Longtin, A., & Seely, A. J. (2011). Review and classification of variability analysis techniques with clinical applications. *Biomedical Engineering Online, 10*, 90.

Breslau, N., Chilcoat, H. D., Kessler, R. C., & Davis, G. C. (1999). Previous exposure to trauma and PTSD effects of subsequent trauma: Results from the Detroit Area Survey of trauma. *American Journal of Psychiatry, 156*(6), 902-907.

Brewin, C. R., Dalgleish, T., & Joseph, S. (1996). A dual representation theory of posttraumatic stress disorder. *Psychological Review, 103*(4), 670.

Brewin, C. R., Gregory, J. D., Lipton, M., & Burgess, N. (2010). Intrusive images in psychological disorders: characteristics, neural mechanisms, and treatment implications. *Psychological Review, 117*(1), 210-232.

Brewin, C. R., & Saunders, J. (2001). The effect of dissociation at encoding on intrusive memories for a stressful film. *British Journal of Medical Psychology, 74*(4), 467-472.

Bryant, R. A., McGrath, C., & Felmingham, K. L. (2013). The roles of noradrenergic and glucocorticoid activation in the development of intrusive memories. *Plos One, 8*, e62675.

Buchanan, T. W., & Lovallo, W. R. (2001). Enhanced memory for emotional material following stress-level cortisol treatment in humans. *Psychoneuroendocrinology, 26*(3), 307-317.

Butler, G., Wells, A., & Dewick, H. (1995). Differential effects of worry and imagery after exposure to a stressful stimulus: A pilot study. *Behavioural and Cognitive Psychotherapy, 23*(1), 45-56.

Cahill, L., Prins, B., Weber, M., & McGaugh, J. L. (1994). β-Adrenergic activation and memory for emotional events. *Nature, 371*(6499), 702-704.

Campolongo, P., Roozendaal, B., Trezza, V., Hauer, D., Schelling, G., McGaugh, J. L., & Cuomo, V. (2009). Endocannabinoids in the rat basolateral amygdala enhance memory consolidation and enable glucocorticoid modulation of memory. *Proceedings of the National Academy of Sciences, 106*(12), 4888-4893.

Chamberlain, S. R., & Robbins, T. W. (2013). Noradrenergic modulation of cognition: therapeutic implications. *Journal of Psychopharmacology, 27*(8), 694-718.

Chapleau, M. W., & Sabharwal, R. (2011). Methods of assessing vagus nerve activity and reflexes. *Heart Failure Review, 16*(2), 109-127.

Charney, D. S., Woods, S. W., Goodman, W. K., & Heninger, G. R. (1987). Neurobiological mechanisms of panic anxiety: biochemical and behavioral correlates of yohimbine-induced panic attacks. *American Journal of Psychiatry, 144*(8), 1030-1036.

Cheung, J., & Bryant, R. A. (2015). FKBP5 risk alleles and the development of intrusive memories. *Neurobiology of Learning and Memory, 125*, 258-264.

Cheung, J., Chervonsky, L., Felmingham, K. L., & Bryant, R. A. (2013). The role of estrogen in intrusive memories. *Neurobiology of Learning and Memory, 106*, 87-94.

Chou, C.-Y., La Marca, R., Steptoe, A., & Brewin, C. R. (2014). Biological responses to trauma and the development of intrusive memories: An analog study with the trauma film paradigm. *Biological Psychology, 103*, 135-143.

Conway, M. A. (2001). Sensory–perceptual episodic memory and its context: autobiographical memory. *Philosophical Transactions of the Royal Society of London. Series B: Biological Sciences, 356*(1413), 1375-1384.

Cornelisse, S., van Ast, V. A., Joëls, M., & Kindt, M. (2014). Delayed effects of cortisol enhance fear memory of trace conditioning. *Psychoneuroendocrinology, 40*, 257-268.

Datson, N. A., van der Perk, J., de Kloet, E. R., & Vreugdenhil, E. (2001). Identification of corticosteroid-responsive genes in rat hippocampus using serial analysis of gene expression. *European Journal of Neuroscience, 14*(4), 675-689.

de Kloet, E. R. (2014). From receptor balance to rational glucocorticoid therapy. *Endocrinology, 155*(8), 2754-2769.

de Kloet, E. R., Joëls, M., & Holsboer, F. (2005). Stress and the brain: from adaptation to disease. *Nature Reviews Neuroscience, 6*(6), 463-475.

de Kloet, E. R., Karst, H., & Joels, M. (2008). Corticosteroid hormones in the central stress response: quick-and-slow. *Frontiers in Neuroendocrinology, 29*(2), 268-272.

de Oliveira Alvares, L., Engelke, D. S., Diehl, F., Scheffer-Teixeira, R., Haubrich, J., de Freitas Cassini, L., . . . Quillfeldt, J. A. (2010). Stress response recruits the hippocampal endocannabinoid system for the modulation of fear memory. *Learning and Memory, 17*(4), 202-209.

de Quervain, D. J., Aerni, A., Schelling, G., & Roozendaal, B. (2009). Glucocorticoids and the regulation of memory in health and disease. *Frontiers in Neuroendocrinology, 30*(3), 358-370.

de Quervain, D. J., Schwabe, L., & Roozendaal, B. (2017). Stress, glucocorticoids and memory: implications for treating fear-related disorders. *Nature Reviews Neuroscience, 18*(1), 7-19.

Dębiec, J., Bush, D. E., & LeDoux, J. E. (2011). Noradrenergic enhancement of reconsolidation in the amygdala impairs extinction of conditioned fear in rats—a possible mechanism for the persistence of traumatic memories in PTSD. *Depression and Anxiety, 28*(3), 186-193.

Delahanty, D. L., Gabert-Quillen, C., Ostrowski, S. A., Nugent, N. R., Fischer, B., Morris, A., . . . Fallon, W. (2013). The efficacy of initial hydrocortisone administration at preventing posttraumatic distress in adult trauma patients: a randomized trial. *CNS Spectrums, 18*(02), 103-111.

Delahanty, D. L., Nugent, N. R., Christopher, N. C., & Walsh, M. (2005). Initial urinary epinephrine and cortisol levels predict acute PTSD symptoms in child trauma victims. *Psychoneuroendocrinology, 30*(2), 121-128.

Diaz-Mataix, L., Piper, W. T., Schiff, H. C., Roberts, C. H., Campese, V. D., Sears, R. M., & LeDoux, J. E. (2017). Characterization of the amplificatory effect of norepinephrine in the acquisition of pavlovian threat associations. *Learning and Memory, 24*(9), 432-439.

Dominique, J.-F., Aerni, A., Schelling, G., & Roozendaal, B. (2009). Glucocorticoids and the regulation of memory in health and disease. *Frontiers in Neuroendocrinology, 30*(3), 358-370.

Ehlers, A. (2010). Understanding and treating unwanted trauma memories in posttraumatic stress disorder. *Journal of Psychology, 218*(2), 141-145.

Ehlers, A., & Clark, D. M. (2000). A cognitive model of posttraumatic stress disorder. *Behaviour Research and Therapy, 38*(4), 319-345.

Ehlers, A., Hackmann, A., & Michael, T. (2004). Intrusive re‐experiencing in post‐traumatic stress disorder: Phenomenology, theory, and therapy. *Memory, 12*(4), 403-415.

Ehlers, A., & Steil, R. (1995). Maintenance of intrusive memories in posttraumatic stress disorder: a cognitive approach. *Behavioural and Cognitive Psychotherapy, 23*(3), 217-249.

Felmingham, K. L., Tran, T. P., Fong, W. C., & Bryant, R. A. (2012). Sex differences in emotional memory consolidation: the effect of stress-induced salivary alpha-amylase and cortisol. *Biological Psychology, 89*(3), 539-544.

Ferree, N. K., & Cahill, L. (2009). Post-event spontaneous intrusive recollections and strength of memory for emotional events in men and women. *Consciousness and Cognition, 18*(1), 126-134.

Ferree, N. K., Kamat, R., & Cahill, L. (2011). Influences of menstrual cycle position and sex hormone levels on spontaneous intrusive recollections following emotional stimuli. *Consciousness and Cognition, 20*(4), 1154-1162.

Finsterwald, C., & Alberini, C. M. (2014). Stress and glucocorticoid receptor-dependent mechanisms in long-term memory: from adaptive responses to psychopathologies. *Neurobiology of Learning and Memory, 112*, 17-29.

First, M., Spitzer, R., Gibbon, M., & Williams, J. B. (1995). *Structured Clinical Interview for DSM-IV Axis I Disorders (SCID)*. New York: New York State Psychiatric Institute, Biometrics Research Department.

Foa, E. B., & Rothbaum, B. O. (2001). *Treating the trauma of rape: Cognitive-behavioral therapy for PTSD*. New York: Guilford Press.

Freund, T. F., Katona, I., & Piomelli, D. (2003). Role of endogenous cannabinoids in synaptic signaling. *Physiologial Reviews, 83*(3), 1017-1066.

Gazarini, L., Stern, C. A. J., Carobrez, A. P., & Bertoglio, L. J. (2013a). Enhanced noradrenergic activity potentiates fear memory consolidation and reconsolidation by differentially recruiting α1-and β-adrenergic receptors. *Learning & Memory, 20*(4), 210-219.

Gazarini, L., Stern, C. A. J., Carobrez, A. P., & Bertoglio, L. J. (2013b). Enhanced noradrenergic activity potentiates fear memory consolidation and reconsolidation by differentially recruiting α1-and β-adrenergic receptors. *Learning and Memory, 20*(4), 210-219.

Geisler, F. C., Kubiak, T., Siewert, K., & Weber, H. (2013). Cardiac vagal tone is associated with social engagement and self-regulation. *Biological Psychology, 93*(2), 279-286.

Gillie, B. L., & Thayer, J. F. (2014). Individual differences in resting heart rate variability and cognitive control in posttraumatic stress disorder. *Frontiers in Psychology, 5*, 758.

Gillie, B. L., Vasey, M. W., & Thayer, J. F. (2014). Heart rate variability predicts control over memory retrieval. *Psychological Science, 25*(2), 458-465.

Gillie, B. L., Vasey, M. W., & Thayer, J. F. (2015). Individual differences in resting heart rate variability moderate thought suppression success. *Psychophysiology, 52*(9), 1149-1160.

Ginsberg, J. P., Berry, M. E., & Powell, D. A. (2010). Cardiac coherence and posttraumatic stress disorder in combat veterans. *Alternative Therapies in Health and Medicine, 16*(4), 52-60.

Giovannitti, J. A., Jr., Thoms, S. M., & Crawford, J. J. (2015). Alpha-2 adrenergic receptor agonists: a review of current clinical applications. *Anesthesia Progress, 62*(1), 31-39.

Graebener, A. H., Michael, T., Holz, E., & Lass-Hennemann, J. (2017). Repeated cortisol administration does not reduce intrusive memories - A double blind placebo controlled experimental study. *European Neuropsychopharmacology, DOI: 10.1016/j.euroneuro.2017.09.001*.

Gurguis, G. N., Vitton, B. J., & Uhde, T. W. (1997). Behavioral, sympathetic and adrenocortical responses to yohimbine in panic disorder patients and normal controls. *Psychiatry Research, 71*(1), 27-39.

Hackmann, A., Ehlers, A., Speckens, A., & Clark, D. M. (2004). Characteristics and content of intrusive memories in PTSD and their changes with treatment. *Journal of Traumatic Stress, 17*(3), 231-240.

Hagenaars, M. A., & Arntz, A. (2012). Reduced intrusion development after post-trauma imagery rescripting; an experimental study. *Journal of Behavior Therapy and Experimental Psychiatry, 43*(2), 808-814.

Halligan, S. L., Michael, T., Clark, D. M., & Ehlers, A. (2003). Posttraumatic stress disorder following assault: the role of cognitive processing, trauma memory, and appraisals. *Journal of Consulting and Clinical Psychology, 71*(3), 419.

Hauer, D., Weis, F., Papassotiropoulos, A., Schmoeckel, M., Beiras-Fernandez, A., Lieke, J., . . . Schelling, G. (2011). Relationship of a common polymorphism of the glucocorticoid receptor gene to traumatic memories and posttraumatic stress disorder in patients after intensive care therapy. *Critical Care Medicine, 39*(4), 643-650.

Hautzinger, M., Bailer, M., Worall, H., & Keller, F. (1995). Beck Depressions Inventar. *Göttingen: Hogrefe*.

Heathers, J. A. (2014). Everything Hertz: methodological issues in short-term frequency-domain HRV. *Frontiers in Physiology, 5*, 177.

Heim, C., & Binder, E. B. (2012). Current research trends in early life stress and depression: Review of human studies on sensitive periods, gene–environment interactions, and epigenetics. *Experimental Neurology, 233*(1), 102-111.

Holmes, E. A., & Bourne, C. (2008). Inducing and modulating intrusive emotional memories: A review of the trauma film paradigm. *Acta Psychologica, 127*(3), 553-566.

Holmes, E. A., Brewin, C. R., & Hennessy, R. G. (2004). Trauma films, information processing, and intrusive memory development. *Journal of Experimental Psychology-General, 133*(1), 3-21.

Holmes, E. A., James, E. L., Kilford, E. J., & Deeprose, C. (2010). Key steps in developing a cognitive vaccine against traumatic flashbacks: visuospatial Tetris versus verbal Pub Quiz. *PLoS One, 5*(11), e13706.

Horowitz, M., Wilner, N., & Alvarez, W. (1979). Impact of Event Scale: a measure of subjective stress. *Psychosomatic Medicine, 41*(3), 209-218.

Insel, T., Cuthbert, B., Garvey, M., Heinssen, R., Pine, D. S., Quinn, K., . . . Wang, P. (2010). Research domain criteria (RDoC): toward a new classification framework for research on mental disorders. *American Journal of Psychiatry, 167*(7), 748-751.

Jacobs, W. J., & Nadel, L. (1985). Stress-induced recovery of fears and phobias. *Psychological Review, 92*(4), 512.

James, E. L., Lau-Zhu, A., Clark, I. A., Visser, R. M., Hagenaars, M. A., & Holmes, E. A. (2016). The trauma film paradigm as an experimental psychopathology model of psychological trauma: intrusive memories and beyond. *Clinical Psychology Review, 47*, 106-142.

Joels, M., & Baram, T. Z. (2009). The neuro-symphony of stress. *Nature Review Neuroscience, 10*(6), 459-466.

Jovanovic, T., & Ressler, K. J. (2010). How the neurocircuitry and genetics of fear inhibition may inform our understanding of PTSD. *American Journal of Psychiatry, 167*(6), 648-662.

Kapfhammer, H. P., Rothenhausler, H. B., Krauseneck, T., Stoll, C., & Schelling, G. (2004). Posttraumatic stress disorder and health-related quality of life in long-term survivors of acute respiratory distress syndrome. *American Journal of Psychiatry, 161*(1), 45-52.

Kemp, A. H., Quintana, D. S., Gray, M. A., Felmingham, K. L., Brown, K., & Gatt, J. M. (2010). Impact of depression and antidepressant treatment on heart rate variability: a review and meta-analysis. *Biological Psychiatry, 67*(11), 1067-1074.

Kessler, R. C., Demler, O., Frank, R. G., Olfson, M., Pincus, H. A., Walters, E. E., . . . Zaslavsky, A. M. (2005). Prevalence and treatment of mental disorders, 1990 to 2003. *The New England Journal of Medicine, 352*(24), 2515-2523.

Kessler, R. C., Sonnega, A., Bromet, E., Hughes, M., & Nelson, C. B. (1995). Posttraumatic stress disorder in the National Comorbidity Survey. *Archives of General Psychiatry, 52*(12), 1048-1060.

Kirschbaum, C., Pirke, K. M., & Hellhammer, D. H. (1993). The 'Trier Social Stress Test' - a tool for investigating psychobiological stress responses in a laboratory setting. *Neuropsychobiology, 28*(1-2), 76-81.

Krauseneck, T., Rothenhausler, H. B., Schelling, G., & Kapfhammer, H. P. (2005). PTSD in somatic disease. *Fortschritte der Neurologie Psychiatrie, 73*(4), 206-217.

Krystal, J. H., & Neumeister, A. (2009). Noradrenergic and serotonergic mechanisms in the neurobiology of posttraumatic stress disorder and resilience. *Brain Research, 1293*, 13-23.

Kuffel, A., Eikelmann, S., Terfehr, K., Mau, G., Kuehl, L. K., Otte, C., . . . Wingenfeld, K. (2014). Noradrenergic blockade and memory in patients with major depression and healthy participants. *Psychoneuroendocrinology, 40*, 86-90.

Kuhlmann, S., & Wolf, O. (2006). Arousal and cortisol interact in modulating memory consolidation in healthy young men. *Behavioral Neuroscience, 120*(1), 217.

Kuhn, E., Blanchard, E. B., Fuse, T., Hickling, E. J., & Broderick, J. (2006). Heart rate of motor vehicle accident survivors in the emergency department, peritraumatic psychological reactions, ASD, and PTSD severity: a 6-month prospective study. *Journal of Trauma Stress, 19*(5), 735-740.

Laborde, S., Mosley, E., & Thayer, J. F. (2017). Heart Rate Variability and Cardiac Vagal Tone in Psychophysiological Research - Recommendations for Experiment Planning, Data Analysis, and Data Reporting. *Frontiers in Psychology, 8*, 213.

Ladwig, K. H., Marten-Mittag, B., Deisenhofer, I., Hofmann, B., Schapperer, J., Weyerbrock, S., . . . Schmitt, C. (2002). Psychophysiological correlates of peritraumatic dissociative responses in survivors of life-threatening cardiac events. *Psychopathology, 35*(4), 241-248.

Laux, L., Glanzmann, P., Schaffner, P., & Spielberger, C. (1981). STAI–Das State-Trait-Angstinventar. *Göttingen, Beltz*.

Lemieux, A. M., & Coe, C. L. (1995). Abuse-related posttraumatic stress disorder: evidence for chronic neuroendocrine activation in women. *Psychosomatic Medicine, 57*(2), 105-115.

Lewis, G. F., Hourani, L., Tueller, S., Kizakevich, P., Bryant, S., Weimer, B., & Strange, L. (2015). Relaxation training assisted by heart rate variability biofeedback: Implication for a military predeployment stress inoculation protocol. *Psychophysiology, 52*(9), 1167-1174.

Lorentz, K., Gütschow, B., & Renner, F. (1999). Evaluation of a direct α-amylase assay using 2-chloro-4-nitrophenyl-α-D-maltotrioside. *Clinical Chemistry and Laboratory Medicine, 37*(11-12), 1053-1062.

Ludäscher, P., Schmahl, C., Feldmann, R. E., Kleindienst, N., Schneider, M., & Bohus, M. (2015). No evidence for differential dose effects of hydrocortisone on intrusive memories in female patients with complex post-traumatic stress disorder–a randomized, double-blind, placebo-controlled, crossover study. *Journal of Psychopharmacology, 29*(10), 1077-1084.

Lupien, S. J., & Lepage, M. (2001). Stress, memory, and the hippocampus: can't live with it, can't live without it. *Behavioural Brain Research, 127*(1-2), 137-158.

Mace, J. (2008). Effects of age on involuntary autobiographical memories. In S. Schlagman, L. Kvavilashvili & J. Schulz (Eds.), *Involuntary Memory* (pp. 87-112). New Jersey: Blackwell Publishing Ltd.

Maercker, A., & Schützwohl, M. (1998). Erfassung von psychischen Belastungsfolgen: Die Impact of Event Skala-revidierte Version. *Diagnostica, 44*(3), 130-141.

Maheu, F. S., Joober, R., Beaulieu, S., & Lupien, S. J. (2004). Differential effects of adrenergic and corticosteroid hormonal systems on human short-and long-term declarative memory for emotionally arousing material. *Behavioral Neuroscience, 118*(2), 420.

Mahon, P. B., Zandi, P. P., Potash, J. B., Nestadt, G., & Wand, G. S. (2013). Genetic association of FKBP5 and CRHR1 with cortisol response to acute psychosocial stress in healthy adults. *Psychopharmacology, 227*(2), 231-241.

Malik, M. (1996). Heart Rate Variability. *Annals of Noninvasive Electrocardiology, 1*(2), 151-181.

McEwen, B. S., Nasca, C., & Gray, J. D. (2016). Stress effects on neuronal structure: hippocampus, amygdala, and prefrontal cortex. *Neuropsychopharmacology, 41*(1), 3-23.

McFarlane, A. C., Barton, C. A., Yehuda, R., & Wittert, G. (2011). Cortisol response to acute trauma and risk of posttraumatic stress disorder. *Psychoneuroendocrinology, 36*(5), 720-727.

McGaugh, J. L. (2004). The amygdala modulates the consolidation of memories of emotionally arousing experiences. *Annual Review of Neuroscience, 27*, 1-28.

McGaugh, J. L. (2013). Making lasting memories: remembering the significant. *Proceedings of the National Academy of Sciences, 110*(2), 10402-10407.

McGaugh, J. L., & Roozendaal, B. (2002). Role of adrenal stress hormones in forming lasting memories in the brain. *Current Opinion in Neurobiology, 12*(2), 205-210.

McGaugh, J. L., & Roozendaal, B. (2009). Drug enhancement of memory consolidation: historical perspective and neurobiological implications. *Psychopharmacology, 202*(1-3), 3-14.

McNally, R. J., Hatch, J. P., Cedillos, E. M., Luethcke, C. A., Baker, M. T., Peterson, A. L., & Litz, B. T. (2011). Does the repressor coping style predict lower posttraumatic stress symptoms? *Military Medicine, 176*(7), 752-756.

Metcalfe, J., & Jacobs, W. (1998). Emotional memory: The effects of stress on "cool" and "hot" memory systems. In D. L. Medin (Ed.), *The psychology of learning and motivation* (Vol. 38, pp. 187-222). New York: Academic Press.

Minassian, A., Maihofer, A. X., Baker, D. G., Nievergelt, C. M., Geyer, M. A., & Risbrough, V. B. (2015). Association of predeployment heart rate variability with risk of postdeployment posttraumatic stress disorder in active-duty marines. *JAMA Psychiatry, 72*(10), 979-986.

Nater, U., & Rohleder, N. (2009). Salivary alpha-amylase as a non-invasive biomarker for the sympathetic nervous system: current state of research. *Psychoneuroendocrinology, 34*(4), 486-496.

Neubig, R. R., Spedding, M., Kenakin, T., & Christopoulos, A. (2003). International Union of Pharmacology Committee on Receptor Nomenclature and Drug

Classification. XXXVIII. Update on terms and symbols in quantitative pharmacology. *Pharmacological Reviews, 55*(4), 597-606.

Nicholson, E. L., Bryant, R. A., & Felmingham, K. L. (2014). Interaction of noradrenaline and cortisol predicts negative intrusive memories in posttraumatic stress disorder. *Neurobiology of Learning and Memory, 112*, 204-211.

Nielsen, S. E., Barber, S. J., Chai, A., Clewett, D. V., & Mather, M. (2015). Sympathetic arousal increases a negative memory bias in young women with low sex hormone levels. *Psychoneuroendocrinology, 62*, 96-106.

Niskanen, J.-P., Tarvainen, M. P., Ranta-Aho, P. O., & Karjalainen, P. A. (2004). Software for advanced HRV analysis. *Computer Methods and Programs in Biomedicine, 76*(1), 73-81.

Nixon, R. D., Bryant, R. A., Moulds, M. L., Felmingham, K. L., & Mastrodomenico, J. A. (2005). Physiological arousal and dissociation in acute trauma victims during trauma narratives. *Journal of Traumatic Stress, 18*(2), 107-113.

O'Carroll, R. E., Drysdale, E., Cahill, L., Shajahan, P., & Ebmeier, K. P. (1999). Stimulation of the noradrenergic system enhances and blockade reduces memory for emotional material in man. *Psychological Medicine, 29*(5), 1083-1088.

O'Donnell, M. L., Elliott, P., Lau, W., & Creamer, M. (2007). PTSD symptom trajectories: from early to chronic response. *Behaviour Research and Therapy, 45*(3), 601-606.

Otte, C., Neylan, T. C., Pole, N., Metzler, T., Best, S., Henn-Haase, C., . . . Marmar, C. R. (2005). Association between childhood trauma and catecholamine response to psychological stress in police academy recruits. *Biological Psychiatry, 57*(1), 27-32.

Owen, J., Nakatsu, S., Fenemore, J., Condra, M., Surridge, D., & Morales, A. (1987). The pharmacokinetics of yohimbine in man. *European journal of clinical pharmacology, 32*(6), 577-582.

Peskind, E. R., Wingerson, D., Murray, S., Pascualy, M., Dobie, D. J., Le Corre, P., . . . Raskind, M. A. (1995). Effects of Alzheimer's disease and normal aging on cerebrospinal fluid norepinephrine responses to yohimbine and clonidine. *Archives of General Psychiatry, 52*(9), 774-782.

Pitman, R. K., & Delahanty, D. L. (2005). Conceptually driven pharmacologic approaches to acute trauma. *CNS Spectrums, 10*(2), 99-106.

Pitman, R. K., Rasmusson, A. M., Koenen, K. C., Shin, L. M., Orr, S. P., Gilbertson, M. W., . . . Liberzon, I. (2012). Biological studies of post-traumatic stress disorder. *Nature Review Neuroscience, 13*(11), 769-787.

Pole, N. (2007). The psychophysiology of posttraumatic stress disorder: a meta-analysis. *Psychological Bulletin, 133*(5), 725-746.

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*(7), 916-931.

Pyne, J. M., Constans, J. I., Wiederhold, M. D., Gibson, D. P., Kimbrell, T., Kramer, T. L., . . . McCune, T. R. (2016). Heart rate variability: Pre-deployment predictor of post-deployment PTSD symptoms. *Biological Psychology, 121*(Pt A), 91-98.

Quas, J. A., Yim, I. S., Rush, E., & Sumaroka, M. (2012). Hypothalamic pituitary adrenal axis and sympathetic activation: Joint predictors of memory in children, adolescents, and adults. *Biological Psychology, 89*(2), 335-341.

Quintana, D. S., & Heathers, J. A. (2014). Considerations in the assessment of heart rate variability in biobehavioral research. *Frontiers in Psychology, 5*, 805.

Quintana, D. S., Heathers, J. A., & Kemp, A. H. (2012). On the validity of using the Polar RS800 heart rate monitor for heart rate variability research. *European Journal of Applied Physiology, 112*(12), 4179-4180.

Quirarte, G. L., Roozendaal, B., & McGaugh, J. L. (1997). Glucocorticoid enhancement of memory storage involves noradrenergic activation in the basolateral amygdala. *Proceedings of the National Academy of Sciences, 94*(25), 14048-14053.

Rajcani, J., Solarikova, P., & Brezina, I. (2017). Increased Heart Rate Variability in Allergic and Highly Trait Anxious Individuals. *Journal of Pharmacy and Pharmacology, 5*, 46-56.

Raskind, M. A., Peskind, E. R., Kanter, E. D., Petrie, E. C., Radant, A., Thompson, C. E., . . . Straits-Tröster, K. (2003). Reduction of nightmares and other PTSD

symptoms in combat veterans by prazosin: a placebo-controlled study. *American Journal of Psychiatry, 160*(2), 371-373.

Regunathan, S., Meeley, M. P., & Reis, D. J. (1991). Clonidine-displacing substance from bovine brain binds to imidazoline receptors and releases catecholamines in adrenal chromaffin cells. *Molecular Pharmacology, 40*(6), 884-888.

Reid, J. L. (1981). The Fourth Lilly Prize Lecture, University of Aberdeen, September 1980. The clinical pharmacology of clonidine and related central antihypertensive agents. *British Journal of Clinical Pharmacology, 12*(3), 295.

Reul, J. M., & de Kloet, E. R. (1985). Two receptor systems for corticosterone in rat brain: microdistribution and differential occupation. *Endocrinology, 117*(6), 2505-2511.

Rodrigues, S. M., LeDoux, J. E., & Sapolsky, R. M. (2009). The influence of stress hormones on fear circuitry. *Annual Review of Neuroscience, 32*, 289-313.

Rombold, F., Wingenfeld, K., Renneberg, B., Hellmann-Regen, J., Otte, C., & Roepke, S. (2016a). Influence of the noradrenergic system on the formation of intrusive memories in women: an experimental approach with a trauma film paradigm. *Psychological Medicine, 46*(12), 2523-2534.

Rombold, F., Wingenfeld, K., Renneberg, B., Schwarzkopf, F., Hellmann-Regen, J., Otte, C., & Roepke, S. (2016b). Impact of exogenous cortisol on the formation of intrusive memories in healthy women. *Journal of Psychiatric Research*.

Rombold-Bruehl, F., Otte, C., Renneberg, B., Bruch, L., Wingenfeld, K., & Roepke, S. (2017a). Impact of stress response systems on forced choice recognition in an experimental trauma film paradigm. *Manuscript submitted for publication*.

Rombold-Bruehl, F., Otte, C., Renneberg, B., Schmied, A., Zimmermann-Viehoff, F., Wingenfeld, K., & Roepke, S. (2017b). Lower heart rate variability at baseline is associated with more consecutive intrusive memories in an experimental distressing film paradigm. *The World Journal of Biological Psychiatry*, DOI: 10.1080/15622975.15622017.11372628.

Roozendaal, B., & McGaugh, J. L. (2011). Memory modulation. *Behavioral Neuroscience, 125*(6), 797.

Roozendaal, B., Okuda, S., de Quervain, D. J., & McGaugh, J. (2006a). Glucocorticoids interact with emotion-induced noradrenergic activation in influencing different memory functions. *Neuroscience, 138*(3), 901-910.

Roozendaal, B., Okuda, S., Van der Zee, E. A., & McGaugh, J. L. (2006b). Glucocorticoid enhancement of memory requires arousal-induced noradrenergic activation in the basolateral amygdala. *Proceedings of the National Academy of Sciences, 103*(17), 6741-6746.

Roozendaal, B., Williams, C. L., & McGaugh, J. L. (1999). Glucocorticoid receptor activation in the rat nucleus of the solitary tract facilitates memory consolidation: involvement of the basolateral amygdala. *European Journal of Neuroscience, 11*(4), 1317-1323.

Rothbaum, B. O., Foa, E. B., Riggs, D. S., Murdock, T., & Walsh, W. (1992). A prospective examination of post-traumatic stress disorder in rape victims. *Journal of Traumatic Stress, 5*(3), 455-475.

Rubin, D. C., Boals, A., & Berntsen, D. (2008). Memory in posttraumatic stress disorder: properties of voluntary and involuntary, traumatic and nontraumatic autobiographical memories in people with and without posttraumatic stress disorder symptoms. *Journal of Experimental Psychology: General, 137*(4), 591-614.

Sammito, S., & Bockelmann, I. (2015). Analysis of heart rate variability. Mathematical description and practical application. *Herz, 40*(1), 76-84.

Sandi, C., & Haller, J. (2015). Stress and the social brain: behavioural effects and neurobiological mechanisms. *Nature Review Neuroscience, 16*(5), 290-304.

Schacter, D. L., Koutstaal, W., & Norman, K. A. (1997). False memories and aging. *Trends in Cognitive Science 1*(6), 229-236.

Schelling, G., Briegel, J., Roozendaal, B., Stoll, C., Rothenhausler, H. B., & Kapfhammer, H. P. (2001). The effect of stress doses of hydrocortisone during septic shock on posttraumatic stress disorder in survivors. *Biological Psychiatry, 50*(12), 978-985.

Schelling, G., Kilger, E., Roozendaal, B., de Quervain, D. J., Briegel, J., Dagge, A., . . . Kapfhammer, H.-P. (2004). Stress doses of hydrocortisone, traumatic

memories, and symptoms of posttraumatic stress disorder in patients after cardiac surgery: a randomized study. *Biological Psychiatry, 55*(6), 627-633.

Schelling, G., Richter, M., Roozendaal, B., Rothenhausler, H. B., Krauseneck, T., Stoll, C., . . . Kapfhammer, H. P. (2003). Exposure to high stress in the intensive care unit may have negative effects on health-related quality-of-life outcomes after cardiac surgery. *Critical Care Medicine, 31*(7), 1971-1980.

Schelling, G., Roozendaal, B., Krauseneck, T., Schmoelz, M., de Quervain, D. J., & Briegel, J. (2006). Efficacy of hydrocortisone in preventing posttraumatic stress disorder following critical illness and major surgery. *Annals of the New York Academy of Sciences, 1071*(1), 46-53.

Schelling, G., Stoll, C., Kapfhammer, H. P., Rothenhausler, H. B., Krauseneck, T., Durst, K., . . . Briegel, J. (1999). The effect of stress doses of hydrocortisone during septic shock on posttraumatic stress disorder and health-related quality of life in survivors. *Critical Care Medicine, 27*(12), 2678-2683.

Schwabe, L., Joëls, M., Roozendaal, B., Wolf, O. T., & Oitzl, M. S. (2012). Stress effects on memory: an update and integration. *Neuroscience & Biobehavioral Reviews, 36*(7), 1740-1749.

Schwabe, L., & Wolf, O. T. (2013). Stress and multiple memory systems: from 'thinking' to 'doing'. *Trends in Cognitive Science 17*(2), 60-68.

Segal, S. K., & Cahill, L. (2009). Endogenous noradrenergic activation and memory for emotional material in men and women. *Psychoneuroendocrinology, 34*(9), 1263-1271.

Segal, S. K., Simon, R., McFarlin, S., Alkire, M., Desai, A., & Cahill, L. (2014). Glucocorticoids interact with noradrenergic activation at encoding to enhance long-term memory for emotional material in women. *Neuroscience, 277*, 267-272.

Segal, S. K., Stark, S. M., Kattan, D., Stark, C. E., & Yassa, M. A. (2012). Norepinephrine-mediated emotional arousal facilitates subsequent pattern separation. *Neurobiology of Learning and Memory, 97*(4), 465-469.

Shalev, A. Y., Peri, T., Canetti, L., & Schreiber, S. (1996). Predictors of PTSD in injured trauma survivors: a prospective study. *American Journal of Psychiatry, 153*(2), 219-225.

Shalev, A. Y., Videlock, E. J., Peleg, T., Segman, R., Pitman, R. K., & Yehuda, R. (2008). Stress hormones and post-traumatic stress disorder in civilian trauma victims: a longitudinal study. Part I: HPA axis responses. *International Journal of Neuropsychopharmacology, 11*(3), 365-372.

Sijbrandij, M., Kleiboer, A., Bisson, J. I., Barbui, C., & Cuijpers, P. (2015). Pharmacological prevention of post-traumatic stress disorder and acute stress disorder: a systematic review and meta-analysis. *The Lancet Psychiatry, 2*(5), 413-421.

Smeets, T., Otgaar, H., Candel, I., & Wolf, O. T. (2008). True or false? Memory is differentially affected by stress-induced cortisol elevations and sympathetic activity at consolidation and retrieval. *Psychoneuroendocrinology, 33*(10), 1378-1386.

Smith, A. L., Owen, H., & Reynolds, K. J. (2013a). Heart rate variability indices for very short-term (30 beat) analysis. Part 1: survey and toolbox. *Journal of Clinical Monitoring and Computing, 27*(5), 569-576.

Smith, A. L., Owen, H., & Reynolds, K. J. (2013b). Heart rate variability indices for very short-term (30 beat) analysis. Part 2: validation. *Journal of Clinical Monitoring and Computing, 27*(5), 577-585.

Sommer, M., Braumann, M., Althoff, T., Backhaus, J., Kordon, A., Junghanns, K., . . . Broocks, A. (2011). Psychological and neuroendocrine responses to social stress and to the administration of the alpha-2-receptor antagonist, yohimbine, in highly trained endurance athletes in comparison to untrained healthy controls. *Pharmacopsychiatry, 44*(4), 129-134.

Southwick, S. M., Bremner, J. D., Rasmusson, A., Morgan, C. A., Arnsten, A., & Charney, D. S. (1999). Role of norepinephrine in the pathophysiology and treatment of posttraumatic stress disorder. *Biological Psychiatry, 46*(9), 1192-1204.

Southwick, S. M., Krystal, J. H., Morgan, C. A., Johnson, D., Nagy, L. M., Nicolaou, A., . . . Charney, D. S. (1993). Abnormal noradrenergic function in posttraumatic stress disorder. *Archives of General Psychiatry, 50*(4), 266-274.

Spielberger, C. D., & Gorsuch, R. L. (1983). *State-Trait Anxiety Inventory for Adults: Manual and Sample: Manual, Instrument and Scoring Guide.* Palo Alto, CA: Consulting Psychologists Press.

Stiglmayr, C. E., Braakmann, D., Haaf, B., Stieglitz, R.-D., & Bohus, M. (2003). Construction and psychometric properties of the Dissoziations-Spannungs-Skala (DSS- akut), a self-rating scale assessing dissociative symptoms and aversive inner tensions. . *Psychotherapy, Psychosomatic, Medical Psychology, 53*(07), 287-294.

Stoll, C., Schelling, G., Goetz, A. E., Kilger, E., Bayer, A., Kapfhammer, H. P., . . . Peter, K. (2000). Health-related quality of life and post-traumatic stress disorder in patients after cardiac surgery and intensive care treatment. *The Journal of Thoracic and Cardiovascular Surgery, 120*(3), 505-512.

Strange, B., Hurlemann, R., & Dolan, R. (2003). An emotion-induced retrograde amnesia in humans is amygdala-and β-adrenergic-dependent. *Proceedings of the National Academy of Sciences, 100*(23), 13626-13631.

Szabo, B., Hedler, L., & Starke, K. (1989). Peripheral presynaptic and central effects of clonidine, yohimbine and rauwolscine on the sympathetic nervous system in rabbits. *Naunyn-Schmiedeberg's Archives of Pharmacology, 340*(6), 648-657.

Takarangi, M. K., Strange, D., & Lindsay, D. S. (2014). Self-report may underestimate trauma intrusions. *Consciousness and Cognition, 27*, 297-305.

Taylor, F. B., Lowe, K., Thompson, C., McFall, M. M., Peskind, E. R., Kanter, E. D., . . . Raskind, M. A. (2006). Daytime prazosin reduces psychological distress to trauma specific cues in civilian trauma posttraumatic stress disorder. *Biological Psychiatry, 59*(7), 577-581.

Tedstone, J. E., & Tarrier, N. (2003). Posttraumatic stress disorder following medical illness and treatment. *Clinical Psychology Review, 23*(3), 409-448.

ter Heegde, F., de Rijk, R. H., & Vinkers, C. H. (2015). The brain mineralocorticoid receptor and stress resilience. *Psychoneuroendocrinology, 52*, 92-110.

Thayer, J. F., Hansen, A. L., Saus-Rose, E., & Johnsen, B. H. (2009). Heart rate variability, prefrontal neural function, and cognitive performance: the neurovisceral integration perspective on self-regulation, adaptation, and health. *Annals of Behavioral Medicine, 37*(2), 141-153.

Thoma, M. V., Joksimovic, L., Kirschbaum, C., Wolf, J. M., & Rohleder, N. (2012). Altered salivary alpha-amylase awakening response in Bosnian War

refugees with posttraumatic stress disorder. *Psychoneuroendocrinology, 37*(6), 810-817.

Toothaker, R. D., Sundaresan, G. M., Hunt, J. P., Goehl, T. J., Rotenberg, K. S., Prasad, V. K., . . . Welling, P. G. (1982). Oral hydrocortisone pharmacokinetics: a comparison of fluorescence and ultraviolet high-pressure liquid chromatographic assays for hydrocortisone in plasma. *Journal of Pharmaceutical Science, 71*(5), 573-576.

Ulrich-Lai, Y. M., & Herman, J. P. (2009). Neural regulation of endocrine and autonomic stress responses. *Nature Review Neuroscience, 10*(6), 397-409.

Urbaniak, G., & Plous, S. (2013). Research Randomizer (Version 4.0)[Computer software].

van Ast, V. A., Cornelisse, S., Meeter, M., Joëls, M., & Kindt, M. (2013). Time-dependent effects of cortisol on the contextualization of emotional memories. *Biological Psychiatry, 74*(11), 809-816.

van der Kolk, B. A., & Fisler, R. (1995). Dissociation and the fragmentary nature of traumatic memories: overview and exploratory study. *Journal of Traumatic Stress, 8*(4), 505-525.

van Rossum, E. F., Binder, E. B., Majer, M., Koper, J. W., Ising, M., Modell, S., . . . Holsboer, F. (2006). Polymorphisms of the glucocorticoid receptor gene and major depression. *Biological Psychiatry, 59*(8), 681-688.

van Rossum, E. F., Koper, J. W., van den Beld, A. W., Uitterlinden, A. G., Arp, P., Ester, W., . . . Lamberts, S. W. (2003). Identification of the BclI polymorphism in the glucocorticoid receptor gene: association with sensitivity to glucocorticoids in vivo and body mass index. *Clinical Endocrinology, 59*(5), 585-592.

van Stegeren, A. (2008). The role of the noradrenergic system in emotional memory. *Acta Psychologica, 127*(3), 532-541.

van Stegeren, A., Goekoop, R., Everaerd, W., Scheltens, P., Barkhof, F., Kuijer, J., & Rombouts, S. A. (2005). Noradrenaline mediates amygdala activation in men and women during encoding of emotional material. *Neuroimage, 24*(3), 898-909.

van Stegeren, A., Rohleder, N., Everaerd, W., & Wolf, O. T. (2006). Salivary alpha amylase as marker for adrenergic activity during stress: effect of betablockade. *Psychoneuroendocrinology, 31*(1), 137-141.

Wallen, M. B., Hasson, D., Theorell, T., Canlon, B., & Osika, W. (2012). Possibilities and limitations of the Polar RS800 in measuring heart rate variability at rest. *European Journal of Applied Physiology, 112*(3), 1153-1165.

Walsh, K., Nugent, N. R., Kotte, A., Amstadter, A. B., Wang, S., Guille, C., . . . Resnick, H. S. (2013). Cortisol at the emergency room rape visit as a predictor of PTSD and depression symptoms over time. *Psychoneuroendocrinology, 38*(11), 2520-2528.

Weidmann, A., Conradi, A., Gröger, K., Fehm, L., & Fydrich, T. (2009). Using stressful films to analyze risk factors for PTSD in analogue experimental studies–which film works best? *Anxiety, Stress, & Coping, 22*(5), 549-569.

Wingenfeld, K., Kuffel, A., Uhlmann, C., Terfehr, K., Schreiner, J., Kuehl, L. K., . . . Spitzer, C. (2013). Effects of noradrenergic stimulation on memory in patients with major depressive disorder. *Stress, 16*(2), 191-201.

Wingenfeld, K., Spitzer, C., Mensebach, C., Grabe, H. J., Hill, A., Gast, U., . . . Driessen, M. (2010). The German version of the Childhood Trauma Questionnaire (CTQ): preliminary psychometric properties. *Psychotherapie, Psychosomatik, Medizinische Psychologie, 60*(11), 442-450.

Wingenfeld, K., Whooley, M. A., Neylan, T. C., Otte, C., & Cohen, B. E. (2015). Effect of current and lifetime posttraumatic stress disorder on 24-h urinary catecholamines and cortisol: Results from the Mind Your Heart Study. *Psychoneuroendocrinology, 52*, 83-91.

Wingenfeld, K., & Wolf, O. (2014a). Stress, memory, and the hippocampus.

Wingenfeld, K., & Wolf, O. (2014b). Stress, memory, and the hippocampus *The Hippocampus in Clinical Neuroscience* (Vol. 34, pp. 109-120): Karger Publisher.

Wingenfeld, K., & Wolf, O. T. (2015). Effects of cortisol on cognition in major depressive disorder, posttraumatic stress disorder and borderline personality disorder - 2014 Curt Richter Award Winner. *Psychoneuroendocrinology, 51*, 282-295.

Wittchen, H.-U., Fydrich, T., & Zaudig, M. (1997). *SKID: Strukturiertes Klinisches Interview für DSM-IV; Achse I und II. Achse I: psychische Störungen. SKID-I*: Hogrefe.

Wittchen, H.-U., Jacobi, F., Rehm, J., Gustavsson, A., Svensson, M., Jönsson, B., . . . Faravelli, C. (2011). The size and burden of mental disorders and other disorders of the brain in Europe 2010. *European Neuropsychopharmacology, 21*(9), 655-679.

Wolf, O. (2009). Stress and memory in humans: twelve years of progress? *Brain Research, 1293*, 142-154.

Wolf, O., Atsak, P., de Quervain, D. J., Roozendaal, B., & Wingenfeld, K. (2016). Stress and memory: A selective review on recent developments in the understanding of stress hormone effects on memory and their clinical relevance. *Journal of Neuroendocrinology, 28*(8), DOI: 10.1111/jne.12353.

Yehuda, R., Hoge, C. W., McFarlane, A. C., Vermetten, E., Lanius, R. A., Nievergelt, C. M., . . . Hyman, S. E. (2015). Post-traumatic stress disorder. *Nature Reviews Disease Primers, 1*.

Zohar, J., Yahalom, H., Kozlovsky, N., Cwikel-Hamzany, S., Matar, M. A., Kaplan, Z., . . . Cohen, H. (2011). High dose hydrocortisone immediately after trauma may alter the trajectory of PTSD: interplay between clinical and animal studies. *European Neuropsychopharmacology, 21*(11), 796-809.

Zoladz, P. R., & Diamond, D. M. (2013). Current status on behavioral and biological markers of PTSD: a search for clarity in a conflicting literature. *Neuroscience & Biobehavioral Reviews, 37*(5), 860-895.

Zucker, T. L., Samuelson, K. W., Muench, F., Greenberg, M. A., & Gevirtz, R. N. (2009). The effects of respiratory sinus arrhythmia biofeedback on heart rate variability and posttraumatic stress disorder symptoms: a pilot study. *Applied Psychophysiology and Biofeedback, 34*(2), 135-143.

Appendix

List of Figures

List of Tables

List of Abbreviations

ACTH - adrenocorticotropin
ANCOVA - Analysis of covariance
ANOVA - Analysis of variance
ANS - Autonomic nervous system
AUC - Area under the curve
AVP - vasopressin
BDI - Beck Depression Inventory
BMI - Body Mass Index
CRF - corticotropin releasing factor
CTQ - Childhood Trauma Questionnaire
DGPs - German Psychology Association
DSS-Acute - The Dissociation-Tension-Scale acute
GABA - Gamma-aminobutyric acid
HPA - Hypothalamus–pituitary–adrenal
HRV - Heart rate variability
IES - Impact of Event Questionnaire
PTSD - Posttraumatic stress disorder
sAA - Salivary alpha-amylase
STAI-T - Trait scale of the Trait Anxiety Inventory
TSST - Trier Social Stress Test
RDoC - Research Domain Criteria

List of Publications

Schultebraucks, K., **Rombold-Bruehl, F.**, Wingenfeld., K., Otte, C., & Roepke, S. (2017). The Impact of acute psychosocial stress on intrusive memories using a trauma film paradigm: A randomized, placebo-controlled study in 120 young females *(in preperation)*.

Rombold-Bruehl F., Otte C., Renneberg B., Bruch L., Wingenfeld K., & Roepke S. (2017). Impact of stress response systems on forced choice recognition in an experimental trauma film paradigm *(under review)*.

Rombold-Bruehl F., Otte C., Renneberg B., Schmied A., Zimmermann-Viehoff, Wingenfeld K, & Roepke S (2017). Lower heart rate variability at baseline is associated with more consecutive intrusive memories in an experimental distressing film paradigm*. The World Journal of Biological Psychiatry*, DOI: 10.1080/15622975.15622017.11372628.
Impact Factor: 3.7

Rombold F, Wingenfeld K, Renneberg B, Schwarzkopf F, Hellmann-Regen J, Otte C*,* *&* Roepke S (2016). Impact of exogenous cortisol on the formation of intrusive memories in healthy women. *Journal of Psychiatric Research* **83**: 71-78.
Impact Factor: 4.2

Rombold F, Wingenfeld K, Renneberg B, Hellmann-Regen J, Otte C, & Roepke S (2016). Influence of the noradrenergic system on the formation of intrusive memories in women: An experimental approach with a trauma film paradigm. *Psychological Medicine* **46**: 2523-2534.
Impact Factor: 5.2

Rombold F, Lauterbach E, Felber W, Mueller-Oerlinghausen B, Ahrens B, Bronisch T, Kilb B, Lewitzka U, Richter K, Broocks A, Heuser I, Hohagen F, & Quante A (2014). Adjunctive lithium treatment in the prevention of suicidal behavior

in patients with depression and comorbid personality disorders. *International Journal of Psychiatry in Clinical Practice* ***18***: 300-303.
Impact Factor: 1.5

Mews MR, **Rombold F**, & Quante A (2014). Efficacy and safety of low-dose doxepin in depressed patients suffering from insomnia: a retrospective, naturalistic case series analysis. *The Primary Care Companion for CNS Disorders* **16**, DOI: 10.4088/PCC.13m01567.
Impact Factor: 1.1

Congress contributions

Talk Wolkenstein L & **Rombold F**. Effects of transcranial direct current stimulation on cognitive control in patients with Borderline Personality Disorder.
35th Annual Conference of the German Psychological Society (DGPs), Chemnitz, 2017

Talk Otte C, **Rombold F**, Wingenfeld K, & Roepke S. Influence of the noradrenergic and glucocorticoid system on the formation of intrusive memories.
46th Annual Conference of the International Society of Psychoneuroendocrinology, Miami, 2016

Talk **Rombold F**, Wingenfeld K, Renneberg B, Hellmann-Regen J, Otte C, & Roepke S. Influence of the noradrenergic system on the formation of intrusive memories: An experimental approach with a trauma film paradigm.
42th Annual conference Psychologie & Gehirn, Berlin, 2016

Poster **Rombold F**, Wingenfeld K, Renneberg B, Hellmann-Regen J, Otte C, & Roepke S. Does the noradrenergic system influence intrusive memories?
45th Annual Conference of the International Society of Psychoneuroendocrinology, Edinburgh, 2015

Poster **Rombold F**, Wingenfeld K, Renneberg B, Hellmann-Regen J, Otte C, & Roepke S. Influence of the noradrenergic system on the formation of intrusive memories: An experimental approach with a trauma film paradigm.
42th Annual conference Psychologie & Gehirn, Berlin, 2016

Curriculum vitae

For data protection reasons, the curriculum vitae is not available in the online version of this dissertation.

Eidesstattliche Versicherung (statement of authorship)

Hiermit versichere ich, dass ich die vorgelegte Arbeit selbstständig verfasst habe und keine anderen als die angegebenen Quellen und Hilfsmittel benutzt wurden. Alle Zitate habe ich kenntlich gemacht.

Die vorliegende Dissertation wurde in keinem vorhergehenden Promotionsverfahren eingereicht und besitze keinen Doktorgrad im Fach Psychologie.

Die Promotionsordnung der Freien Universität Berlin vom 27.10.1998, zuletzt geändert am 08.08.2016 und veröffentlicht im Amtlichen Mitteilungsblatt Nr. 35/2016, ist mir bekannt.

Berlin, den 17. Dezember 2017

_____

Felicitas Rombold-Brühl