A MULTIMODAL INVESTIGATION OF STRESS EFFECTS ON MEMORY PROCESSES

by

Nadja Herten

A thesis submitted in partial fulfilment of the requirements for the degree of

Philosophiae Doctoris (PhD) in Neuroscience

From the International Graduate School of Neuroscience

Ruhr University Bochum

January 16th 2017

This research was conducted at the Department of Cognitive Psychology, within the Faculty of Psychology at the Ruhr University under the supervision of Prof. Dr. Oliver T. Wolf

Printed with the permission of the International Graduate School of Neuroscience, Ruhr University Bochum

PhD Commission

Chair:

1st Internal Examiner: Prof. Dr. Oliver T. Wolf

2nd Internal Examiner: Prof. Dr. Martin Brüne

External Examiner:

Non-Specialist:

Date of Final Examination:

PhD Grade Assigned:

Statement

I certify herewith that the dissertation included here was completed and written independently by me and without outside assistance. References to the work and theories of others have been cited and acknowledged completely and correctly. The "Guidelines for Good Scientific Practice" according to § 9, Sec. 3 of the PhD regulations of the International Graduate School of Neuroscience were adhered to. This work has never been submitted in this, or a similar form, at this or any other domestic or foreign institution of higher learning as a dissertation.

The abovementioned statement was made as a solemn declaration. I conscientiously believe and state it to be true and declare that it is of the same legal significance and value as if it were made under oath.

Nadja Herten

Bochum, 16.01.2017

Table of contents

I. List of Figures

II. List of Tables

III. List of Abbreviations

ANCOVA	analyses of covariance
ANOVA	analyses of variance
AOI	Area of Interest
AUC_i	Area Under the Curve regarding increase
BMI	body mass index
CPT	Cold Pressor Task
CV	coefficient of variation
DELFIA	Dissociation-Enhanced Lanthanide Fluorescent Immunoassay
DI	discrimination index
EC	entorhinal cortex
ELISA	enzyme-linked immunosorbent assay
EMG	electromyography
ERP	event-related potential
fMRI	functional magnetic resonance imaging
f-TSST	friendly Trier Social Stress Test
GR	glucocorticoid receptor
HPA	hypothalamus-pituitary-adrenal
IAPS	International Affective Picture System
ISI	inter-stimulus interval
LTP	long-term potentiation
MR	mineralocorticoid receptor
NA	negative affect
NEO-FFI	Neuroticism, Extraversion, Openness for experience - Five Factors Inventory
PA	positive affect
PANAS	Positive and Negative Affect Scale
PET	positron emission tomography
PFC	prefrontal cortex
PTSD	Post-Traumatic Stress Disorder
sAA	salivary α-amylase
SD	standard deviation
SECPT	Socially Evaluated Cold Pressor Task
SIAS	Social Interaction Anxiety Scale

SNS	sympathetic nervous system
TSST	Trier Social Stress Test

IV. Abstract

The organism's response to stress is an adaptive mechanism for keeping up the homeostasis in the moment of stress as well as for preparing the organism for similar situations reoccurring in the future. The stress response thus exerts influence on attentional and memory encoding processes from the onset of the stressful situation and in its aftermath when consolidation processes come into play. As a result, memory for aspects and objects of a stressful episode has been found to be enhanced in comparison to a non-stressful experience. This was shown to be particularly pronounced for central, in some way meaningful, aspects of the stressful situation. The underlying mechanism which has been made responsible for this is co-activation of the sympathetic nervous system and hypothalamus-pituitary-adrenal axis, resulting in increased cortisol release. These neuroendocrine influences act on brain areas involved in memory processes. It is yet unclear whether stress also modifies fixation and hereby attentional processes which, in concert with the psychophysiological effects, lead to memory enhancement. Experiment 1 therefore aimed at investigating fixation behaviour under stress by means of a mobile eye tracking device. While participants randomly took part in the Trier Social Stress Test (TSST), holding a free speech in front of an evaluation committee, or the friendly control condition (f-TSST), both including 20 office items, their fixations were recorded with the eye tracker. The committee used 10 of the items whereby these became central. One day later, the participants' memory for the items was tested with free recall and object recognition tasks. It was shown that stressed participants exhibit more and longer fixations on the central items and also show better memory for these. However, memory and fixation measures did not correlate, nor did fixation behaviour mediate memory outcome in the stress group. Fixation on the faces of the committee members was reversed, with participants from the control group fixating the faces longer and more often than participants stressed. Experiment 1 demonstrates that stress influences fixation behaviour towards longer and more fixations on objects related to the stressful episode. Nevertheless, no direct translation of fixation into memory measures could be shown.

Olfactory stimuli have been shown to play a special role in emotional memory processes. Due to the connection of the olfactory system with amygdaloidal structures, odours are prone to be related to affective and personal memories. Stress is thus likely to even enhance the strong emotional component of an olfactory experience leading to a solid memory trace. Since stress leads to increased vigilance, the responsivity to intense stimuli in any sensory modality gets more pronounced. The human auditory startle eye-blink response, evoked by a loud and instant white noise, has been shown to get potentiated by emotionally laden stimuli (in any

modality), but has not yet been investigated in combination with stress induction via the TSST. Experiment 2 was thus designed to assess the impact of stress, inducing increased vigilance, on startle responsivity 24 hours later, with special focus on odour memory. Therefore, an unknown and neutral odour was dispersed in the room where the TSST/f-TSST took place which one day later was re-experienced during an auditory startle session via olfactometer. It was shown that stressed participants exhibit generally enhanced startle responsivity, whereas specificity in response to the odour ambient in the TSST room on the previous day was found to be decreased. Increased vigilance under stress might lead to a shift in amygdaloidal functioning towards enhanced responsivity at the expense of a differential response. Moreover, explicit memory for the odour was poor and did not differ between stress and control group. Participants of the stress group however rated the odour more negative than control participants, at a trend level. Experiment 2 was the first to demonstrate a stress-induced increase in auditory startle responsivity 24 hours after psychosocial stress exposure.

Since memory enhancement for aspects of a stressful episode has mainly been investigated in terms of long-term effects, at least one day after stress exposure, experiment 3 aimed at expanding the findings to a short delay between stress and memory assessment. Additionally, the effects of acutely enhanced vigilance on startle responsivity and response specificity combined with olfactory memory were assessed to expand the findings of experiment 2. With the same methods as in the previous experiments, participants were randomly exposed to psychosocial stress. Office items and an ambient odour were present in the testing room, and recognition and free recall were tested after a startle block featuring the odour experienced during stress as well as distractor odours. Results of the experiment could show that the stress effects on memory are present already in immediate aftermath of the stressor and thus do not mainly rely on consolidation processes. The increased vigilance was shown to enhance the startle responsivity. At a trend level also startle specificity in response to the ambient odour was found to be more pronounced in stressed participants. Whereas memory for the odour tended to be better in the stress group, the odour ratings did not differ between stress and control group, in contrast to experiment 2. Experiment 3 showed acute stress effects on memory enhancement and increased startle responsivity.

This dissertation demonstrates that stress effects on memory occur immediately, reflected in fixation differences during encoding as well as in shortly delayed memory enhancement, in addition to better memory performance after one day. Furthermore, increased vigilance was found to enhance the human startle response and interestingly do so even 24 hours after the one-time stress experience.

1. General introduction

Research has contributed a lot to understanding of how stress can be beneficial for memory. Light has been shed on the interaction of brain regions such as amygdala, hippocampus, and prefrontal cortex and the involvement of hormones and neurotransmitters during the stress response. Knowledge about the importance of timing in between stressor, encoding of to be learned material or acquisition, retrieval, and extinction could be applied to therapy of fear related diseases and illnesses such as Post-Traumatic Stress Disorder (PTSD). However, the aforementioned processes only work as good as our senses perceiving stimuli and noticing threat which is the initiation of a cascade of reactions our organism has in store to cope with the situation at hand. Threat to the organism's homeostasis may be noticed by our olfactory system, for instance by perceiving the smell of fire. Our auditory system may also be the first to send out a warning to our finely tuned response network after having heard a loud noise such as an explosion. Often, our visual sense is the dominating source of information about a potential danger. Most of the time, though, it is an interplay of the perception all our senses provide us with in combined force to react to any kind of threat as quickly and adequately as possible. As described here, threat to our organism may come as a sudden danger. For the majority of us it is a continuous process that disequilibrates our balance, most of the time by constant job stress and the difficulty to maintain a work-life balance in the modern meritocracy. For keeping up our balance, it is useful to understand the influence of stress on our perception and attentional processes as well as on our higher cognitive functions such as memory. As our senses altogether contribute to providing our brain with information which is the initial point of dealing with a stressful situation, I with this dissertation aim at establishing a link between stress, visual and olfactory perception, and memory outcome. The main questions of the studies forming the groundwork to this dissertation are: Does stress exert an influence on fixation behaviour, hereby accounting for enhanced memory performance? Can a past stressful experience be assessed pre-attentively by applying an auditory startle paradigm 24 hours later? May re-exposure to an olfactory stimulus experienced during a stressful episode trigger a fear-potentiated startle response 24 hours later? Is there a difference between a delayed and an immediate assessment during the acute stress response? Can a one-time stress exposure involving an ambient odour lead to a more aversive rating of this odour in the immediate or distant aftermath and enhance its recognition?

1.1. Stress

Despite being a term with mainly negative associations, stress is a well-coordinated response of our organism to any kind of threat to its homeostasis. Hence, it is an adaptive coping strategy, making maximum use of the sources available (McEwen, 1998) – a process termed "allostasis" (Sterling & Eyer, 1988).

The stress response proceeds via two main pathways responsible for maintenance of the homeostasis. The first one is a fast acting response chain initiated by activation of the main division of the autonomous nervous system, the sympathetic nervous system (SNS; *Figure 1.1.a*).

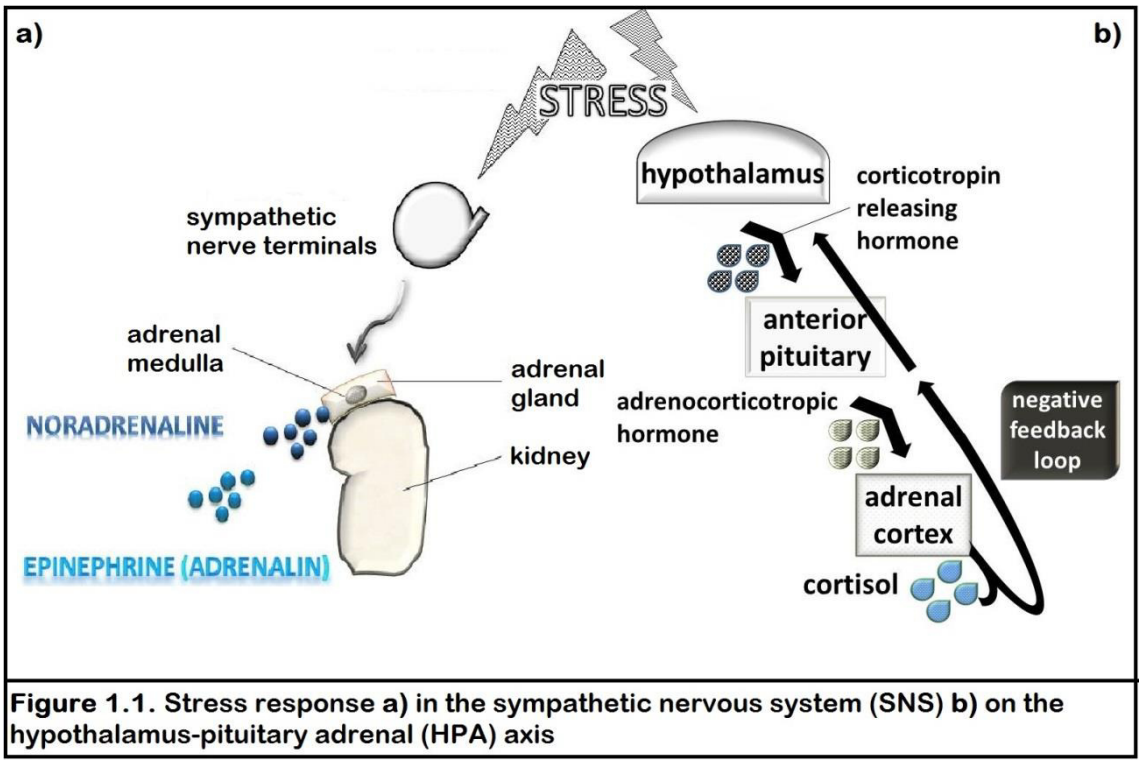

Figure 1.1. Stress response a) in the sympathetic nervous system (SNS) b) on the hypothalamus-pituitary adrenal (HPA) axis

Release of the hormone and neurotransmitter noradrenaline, a catecholamine produced in the locus coeruleus (Fuxe et al., 1970), from sympathetic nerve terminals, and adrenalin release from the adrenal medulla initiate a quick "fight-or-flight" reaction (Cannon, 1913; Goldstein, 2003). The typical physiological symptoms evoked by this include increased heart rate and blood pressure, pupil dilation, and overall enhanced vigilance and alertness, promoting focussed attention and energy mobilisation (de Kloet, Joëls, & Holsboer, 2005). Since release of the enzyme salivary α-amylase (sAA; a-1,4-a-D-glucan 4-glucanohydrolase; EC 3.2.1.1) is mainly stimulated by the neurotransmitters noradrenaline and epinephrine, it can serve as a

biomarker for activation of the SNS (Nater & Rohleder, 2009). The stress response of the SNS is terminated by activation of the parasympathetic nervous system after the threat is over (Ulrich-Lai & Herman, 2009). The second autonomic response cascade to stress-induced emotional arousal proceeds via the hypothalamus-pituitary-adrenal (HPA) axis and is slower acting (Ulrich-Lai & Herman, 2009; *Figure 1.1.b*). By activation of this pathway, neurons of the paraventricular nucleus of the hypothalamus secrete corticotropin-releasing hormone and arginine vasopressin, promoting adrenocorticotropic hormone release from the anterior pituitary. This leads to secretion of glucocorticoids, which in humans is the hormone cortisol, by the adrenal cortex. Glucocorticoids initiate a negative feedback-loop to mark the termination of the stress response (Ulrich-Lai & Herman, 2009). They bind to glucocorticoid (GR) as well as mineralocorticoid receptors (MR), both in numerous expressions in hippocampus (Herman, Patel, Akil, & Watson, 1989; van Steensel et al., 1996). In other brain regions GRs are considerably predominant and widespread (de Kloet, Oitzl, & Joëls, 1999). The affinity of GRs for corticosterone and cortisol is much lower than the case for the MRs, having a 10-fold higher affinity, such that they are occupied even under non-stressful conditions (Reul & de Kloet, 1985). Full activation of GRs is achieved through high levels of cortisol. Thus, MR activation causes utterly different response patterns than co-activation of both receptor types (de Kloet et al., 1999) – an attribute determining the characteristics of the stress response. A high density of MR and GR receptors in prefrontal cortex is further responsible for a quick and flexible response to the specific stressful situation at hand. Top-down control based goal-directed behaviour relying mainly on prefrontal areas is impaired, whereas rather habitual striatum based behaviour is promoted (Schwabe & Wolf, 2010). These processes elicit instant habitual responding based on experience rather than time inefficient top-down problem solving behaviour.

1.1.1. Measuring stress by salivary cortisol

For assessing the stress response in terms of HPA axis activation in experiments, cortisol is a convenient candidate. Above all, sampling can be done collecting saliva, which is a simple and minimally invasive method (Hellhammer, Wüst, & Kudielka, 2009; Kirschbaum & Hellhammer, 1989). Additionally, cortisol can also be extracted from blood, urine or hair samples. When taking full consideration of its characteristics, salivary cortisol is a reliable tool to assess the stress response. Therefore, cortisol's circadian rhythm (Debono et al., 2009; Kirschbaum & Hellhammer, 1989) has to be taken into account - a strong cortisol awakening response in the morning with a steady decline within the course of the day (Fries, Dettenborn,

& Kirschbaum, 2009), causing baseline variation dependant on the time of day. Further factors that lead to a generally lower cortisol level are intake of hormonal contraceptives (Nielsen, Segal, Worden, Yim, & Cahill, 2013) as well as excessive sports, whereas higher cortisol release is caused by obesity (Björntorp & Rosmond, 2000), aging, and consumption of alcohol (Badrick et al., 2008), nicotine (Kirschbaum, Wüst, & Strasburger, 1992) and other forms of medication operating via components of the HPA axis (Hellhammer et al., 2009; Kudielka, Hellhammer, & Wüst, 2009). When testing female participants it has to be considered that cortisol release varies with menstrual cycle phases; whereas during the luteal phase the cortisol response in women is comparable to that in men (Schoofs & Wolf, 2009), women show a blunted response during follicular phase and menses and when taking hormonal contraceptives (Kirschbaum, Kudielka, Gaab, Schommer, & Hellhammer, 1999). In addition, women's cortisol response to certain stressors has sometimes been shown to be generally less pronounced than men's (Kirschbaum et al., 1999).

1.1.2. Laboratory stressors

Various forms of stressors have been used to induce a stress response in the laboratory. Since physiological threat to the homeostasis also causes the organism to respond with stress, that is autonomic activation (e. g. heart rate increase, pupil dilation) as well as cortisol elevation, it is a very simple method to set the organism's parameters out of balance, for instance by affecting body temperature. The Cold Pressor Task (CPT) therefore is a very common laboratory stressor, consisting of the sheer physiological response of resting a hand in ice cold water. As psychosocial stress was found to elicit the strongest stress responses (Skoluda et al., 2015), the CPT was modified adding a psychosocial component as Socially Evaluated CPT (SECPT; Schwabe, Haddad, & Schachinger, 2008). Hereby, the described procedure is videotaped by a reserved experimenter watching the participant and later reportedly analysing their facial expressions. Even though the SECPT includes both physiological as well as psychosocial components, the Trier Social Stress Test (TSST; Kirschbaum, Pirke, & Hellhammer, 1993) was found to be the strongest laboratory stressor (Dickerson & Kemeny, 2004; Skoluda et al., 2015). It elicits a strong and reliable cortisol response typically exhibiting a peak approximately 10 minutes after termination of the stressor (Kirschbaum et al., 1993). In the paradigm, the participant is being led into a room with an evaluation committee introduced as behavioural psychologists analysing the participants' behaviour during a free speech. The situation is videotaped and the speech is held in front of a microphone, supposedly for further analysis of both video and audio parameters. The

participant is instructed to imagine applying for a job while only referring to their personality traits qualifying them for the respective job position. In the original version, participants are allowed a five minutes preparation phase followed by another five minutes of speech, and an arithmetic task of counting backwards from 2043 in steps of 17 for another 5 minutes. If participants make a mistake in the arithmetic task, they have to start again. The features contributing to the TSST being the strongest stressor are uncontrollability, external social evaluation, and motivation to succeed (Dickerson & Kemeny, 2004; Skoluda et al., 2015). Participants experience a situation of threat to their self-esteem which they cannot fully influence. In the TSST, this is achieved by an evaluation committee wearing white laboratory coats and not showing any reaction to the participants' performance, such that the participant lacks feedback, which is likely to evoke a feeling of failure. The two committee members further take notes and direct gaze to the participant during their speech to elicit the feeling of being socially evaluated. Since success in applying for a job is a common pursuit, participants naturally are intrinsically motivated to perform well. By videotaping the situation such that the participant can see themselves on a screen, the feeling of being socially evaluated is increased, further enhancing stress. By instructing the participant to only refer to their personal skills and character traits qualifying them for the respective job position, the speech contents are restricted. If not a trained speaker, the participant thus usually talks for only a few minutes about their personality before stopping or starting to refer to qualifications achieved or curriculum vitae. It is then that the committee interrupts the participant and reminds them to only refer to their personality and character traits. If not spoken for longer than 20 seconds, the committee asks the participant to resume their speech by saying that there is still time left. Participants are never told about how much time they have for their speech. Questions are not answered by the committee members.

1.2. Stress and memory

The function of the stress response is not restricted to an instant reaction to threat for keeping up the homeostasis, but additionally has a future-oriented component. That is, to remember the circumstances, but above all the most crucial stimuli of the stressful experience as precisely as possible for similar situations occurring in the future (de Kloet et al., 2005). Hence, the stress response exerts a pivotal influence on memory formation processes from the moment of stress onset and still enduring in the aftermath of a stressful situation.

Memory formation is a continuous process, as we constantly need to keep things in mind across short time spans while we are working and during all other different forms of

behaviour. In our working memory, part of the short-term memory (*see for review*: Baddeley, 2003), memory contents are still accessible and can be manipulated, making it an extreme form of short-term memory which describes temporary storage of information (*see for review*: Baddeley, 2012). However, to store the temporary information needed in a particular situation for an endless amount of time would not be efficient and cognitively economic. Thus, only selective memory contents are transferred into long-term memory storage, depending on relevance, salience, and rehearsal (e. g. Atkinson & Shiffrin, 1968; Smeets et al., 2009; Sutherland & Mather, 2012). Whereas short-term memory relies on structures in prefrontal cortex, long-term memory consists of the medial temporal lobe system with hippocampus, entorhinal, perirhinal, and parahippocampal cortices (hippocampal region; Baddeley, 2001).

But what aspects does it depend on whether information gets consolidated into long-term memory? A crucial part of this decision depends on limbic areas, especially on activation of the basolateral complex of the amygdala during memory encoding (Roozendaal, McEwen, & Chattarji, 2009; Roozendaal & McGaugh, 2011). Memory encoding is the phase of learning or acquisition of memory, consolidation is when the encoded information is being transferred into long-term memory for permanent storage, and retrieval is recall or recognition of stored memory content. The influence of stress on memory processes depends on the timing of the stressor, precisely on whether the stressor occurs during encoding, consolidation or retrieval (Joëls, Pu, Wiegert, Oitzl, & Krugers, 2006; Roozendaal, 2002; Roozendaal & McGaugh, 2011). While stress and arousal are beneficial for encoding and consolidation (Henckens et al., 2009; Joëls et al., 2006; Lupien et al., 2002) to enable remembering the most crucial aspects of a stressful situation for the future, it impedes or even utterly blocks memory retrieval (de Quervain, Roozendaal, & McGaugh, 1998; de Quervain, Roozendaal, Nitsch, McGaugh, & Hock, 2000). This effect, in real-life situations such as oral examinations sometimes experienced as “blackout”, is apparently due to cortisol causing a consolidation state of the brain while impairing retrieval (Kuhlmann & Wolf, 2006). As described in section 1.1., the allostasis causes the organism to switch to a state of maximum efficiency, also reflected by a shift of instrumental behaviour from more prefrontal cortex based goal-directed to dorsolateral striatum based habitual behaviour (Schwabe, Tegenthoff, Hoffken, & Wolf, 2010), while executive control is repressed (Elzinga & Roelofs, 2005). This demonstrates the focus of the stress response on optimum use of the available sources. Additionally, an optimal state for neuronal plasticity induced by stress hormones is created (Henckens et al., 2009). Both, the effect of stress and emotion in the phase of encoding as well as during the consolidation processes after encoding are mediated by amygdala activation (Hamann, 2001).

During acute stress, locus coeruleus, core of the SNS, floods hippocampus and amygdala with noradrenaline, accounting for a hypervigilant state which lasts for approximately half an hour (Joëls, Fernandez, & Roozendaal, 2011), interacting with neuromodulators like acetylcholine and stress hormones for long-term memory formation (McGaugh & Roozendaal, 2002, 2009). A low-affinity membrane version of MRs is involved in quick (several minutes) and reversible non-genomic actions of glucocorticoids in hippocampus (de Kloet, Karst, & Joëls, 2008; Joëls, Karst, DeRijk, & de Kloet, 2008), which in neurons of basolateral amygdala can have long-lasting encoding effects for emotional stimuli (Karst, Berger, Erdmann, Schütz, & Joëls, 2010). During this phase, long-term potentiation (LTP) is increasingly promoted (Wiegert, 2006). Hereby, the amygdala functions as a neuronal interface integrating actions of noradrenergic processes and stress hormones to facilitate memory consolidation (Roozendaal, 2000). In addition, the basolateral amygdala is equipped with corticosteroid receptors, such that there is not only an indirect, but also a direct influence of stress hormones on amygdala activation (Kim & Diamond, 2002; Reul & de Kloet, 1985). The influence cortisol exerts on noradrenergic action in the nucleus of the basolateral amygdala is crucial for memory enhancement; in turn amygdala activation is a prerequisite for cortisol's effects on memory (Roozendaal, 2000). Slower (≈ 1 hour) genomic actions involve both receptor types, MRs and GRs, and induce long-lasting changes and memory traces if the encoded material is crucial enough, as during this phase, the potential for LTP induction is reduced (Kim & Diamond, 2002). These processes reflect that combined action of the HPA-axis and the SNS is a prerequisite for the beneficial effects of stress on memory. Amygdala activation hereby leads to a superiority for emotional information to get stored and also consolidated into long-term memory (Abercrombie, Speck, & Monticelli, 2006; Cahill et al., 1996; Canli, Zhao, Brewer, Gabrieli, & Cahill, 2000). Emotional memory contents are better remembered, even in absence of enhanced cortisol release during encoding or consolidation (Abercrombie, Kalin, Thurow, Rosenkranz, & Davidson, 2003). This effect can partly be attributed to emotional binding, an item-specific consolidation process for storage of item-emotion bindings by regions in the medial temporal lobe, mediated by the amygdala (Mather, 2007; Yonelinas & Ritchey, 2015). Enhancing effects of emotional, in particular highly arousing and stressor-related, stimuli on memory have been found to be facilitated by stress (Smeets et al., 2009). Hereby, valence seems to be not as important a determinant for amygdala activation (Canli et al., 2000; Hamann, Ely, Grafton, & Kilts, 1999) and memory outcome as arousal. While cortisol release is not necessarily related to enhanced subjectively experienced negative affect, subjective emotional intensity seems to correlate well with amygdala activation (Canli et al.,

2000). It is suggested that it contributes to affective responses such that emotional arousal is actually the outcome of amygdala activation (Canli et al., 2000). As not mentioned so far, the amygdala has several direct as well as indirect connections to hippocampal regions (Pikkarainen, Rönkkö, Savander, Insausti, & Pitkänen, 1999) via which it exerts crucial influence on hippocampal functioning (*see for review*: Kim & Diamond, 2002). Evidence shows that co-activation of hippocampus and amygdala, combined with release of the neuromodulators involved in the stress response, are a prerequisite for memory effects, and that the amygdala plays a central role in stress effects on memory encoding and consolidation (Cahill & McGaugh, 1998; Hamann, 2001; LeDoux, 2000). Additionally, amygdala activity during encoding can predict memory outcome (Canli et al., 2000; Hamann et al., 1999), even so in a linear relation – the stronger the amygdala is activated, the higher the potency of glucocorticoids to enhance memory for emotional input (Canli et al., 2000). Since one of the main input channels is our visual system, it is likely to be influenced by stress leading to altered fixation and attention processes.

Memory enhancement under stress has been shown to be initiated from the moment of stress onset with enhanced attentional selectivity (Chajut & Algom, 2003) for the potentially meaningful items competing to be encoded. Different attentional processes under stress are responsible for this effect. Evidence indicates that stress contributes to peripheral narrowing (Williams, Tonymon, & Andersen, 1991). This shows a stress-induced modification of visual processes, reflecting an effect of stress at an early perceptual stage. In situations with socially induced stress, the narrowing of focus is even more pronounced (*see for review*: Chajut & Algom, 2003). Thus, stress induction with the TSST is well-suited for investigations on these effects in combination with memory performance. It has previously been demonstrated that memory for objects in the TSST is enhanced when assessed 24 hours later (Wiemers, Sauvage, Schoofs, Hamacher-Dang, & Wolf, 2013). Since this effect was particularly prominent for objects which have been central for the situation, this suggests that attentional narrowing processes had taken place. That is, attention is drawn towards potentially relevant and salient items *(see for review:* Christianson, 1992). However, fixation behaviour as a measure of attention towards items in a stressful situation such as the TSST has not been investigated yet.

1.3. Olfaction

As described in the previous section, the encoding of stimuli depends on the arousing or stressful context of the situation, as of relevance for the current thesis, as well as on features

of the to be encoded stimuli themselves. Whereas emotionally laden objects in themselves lead to more pronounced encoding, even more enhanced in an emotional or stressful situation, other objects might only gain relevance through an emotional/stressful context. Besides these effects, stimuli can have a special potency of being well encoded and remembered due to the modality they are presented in. Since a special potency of stimuli presented in the olfactory modality with regard to memory processes has been believed and discussed since more than a century, part of this thesis is dedicated to the role of olfactory stimuli in memory formation under stress.

Already in the 18th century, Schultze (1863) provided accurate description and graphical illustration of olfactory receptor cells (Zippel, 1993). Receiving early attention by scientists and researchers, our olfactory system in fact holds a special role amongst our senses which is mainly owed to the close connectivity of the olfactory system with emotional structures. The olfactory system consists of the olfactory bulb, the olfactory epithelium, which is part of the olfactory mucosa (Doty, 2015), and the olfactory cortex with anterior olfactory nucleus, tenia tecta, olfactory tubercle, piriform cortex, lateral entorhinal cortex, periamygdaloid cortex, and cortical nucleus of the amygdala (Doty, 2001; *Figure 1.2.*, partly based on Wilson, Chapuis, & Sullivan, 2015). Right orbitofrontal cortex was found to be activated by odours together with the piriform cortex (Zatorre, Jones-Gotman, Evans, & Meyer, 1992) which it has a strong reciprocal connection with (Ekstrand et al., 2001; Johnson & Leon, 2000). Orbitofrontal cortex is not exclusively dedicated to the olfactory system, but involved in activity within all sensory systems.

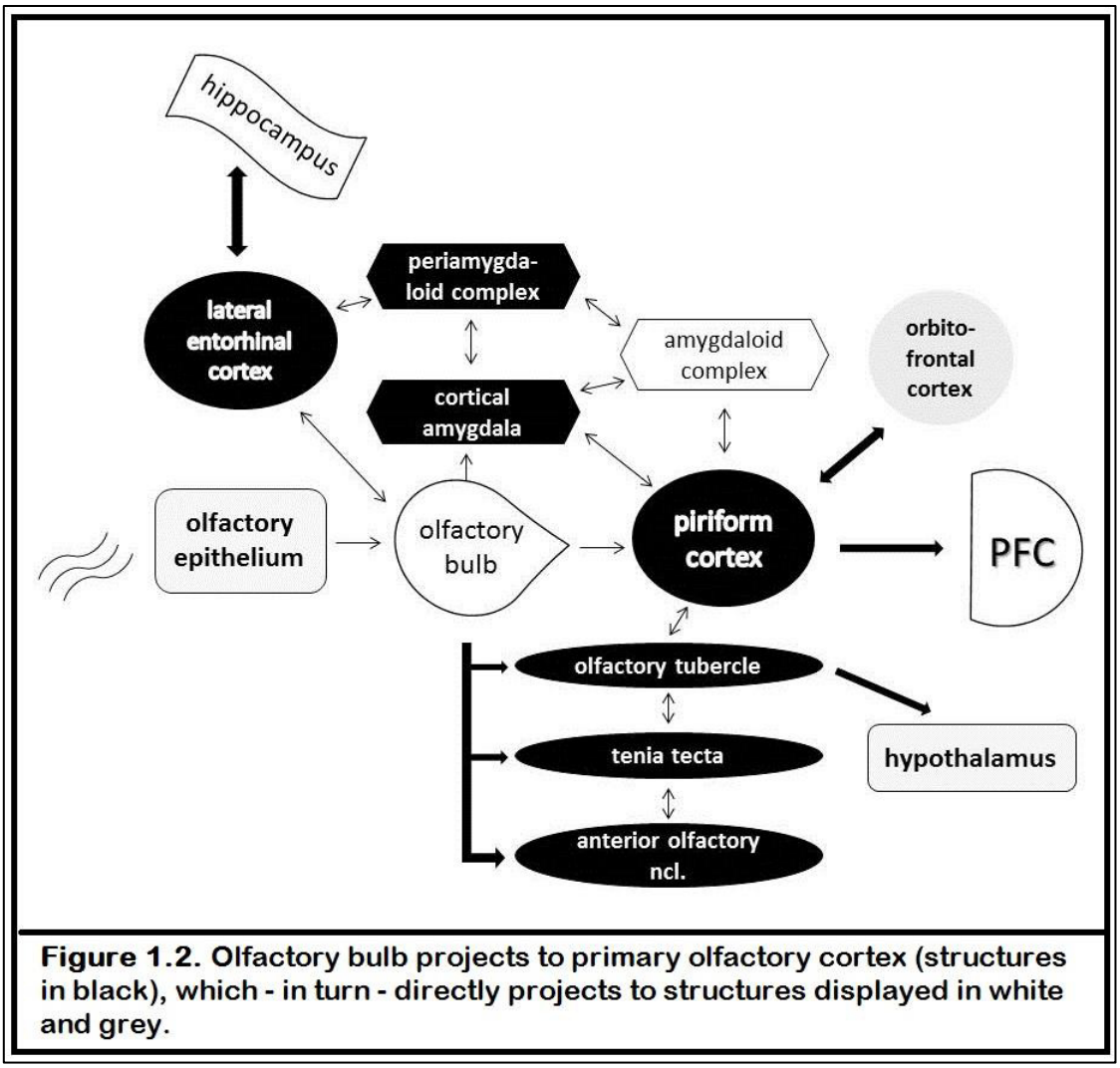

Figure 1.2. Olfactory bulb projects to primary olfactory cortex (structures in black), which - in turn - directly projects to structures displayed in white and grey.

The olfactory cortex fulfils the role of primary sensory and association cortices, such that both characteristic as well as context information of the odour are being integrated (Gottfried, 2009, 2010). The olfactory bulb consists of mitral cells whose axons form the lateral olfactory tract and in form of a fibre bundle project to the "vomeronasal amygdala" which consists of (postero)medial cortical nuclei of the amygdala and the nucleus of the accessory olfactory tract (Kevetter & Winans, 1981). By means of glomeruli, the mitral cells communicate with the bipolar receptor cells via their axon terminals (Doty, 2001). Lateral entorhinal cortex receives input from mitral cells and piriform cortex (Carmichael, Clugnet, & Price, 1994). Its cell units express more response selectivity than cells in the rest of the system (Xu & Wilson, 2012).

Volatile odours are perceived by the olfactory epithelium where olfactory receptor neurons synapse in the main olfactory bulb (Ennis & Holy, 2015). In contrast to neurons of other sensory modalities, neurons in the olfactory epithelium are able to regenerate after damage

(Jang, Youngentob, & Schwob, 2003; Leung, Coulombe, & Reed, 2007; Iwai, Zhou, Roop, & Behringer, 2008), even in older people (Holbrook, Wu, Curry, Lin, & Schwob, 2011), which was discovered in 1940 in rodents and 20 years later in primates (Schultz, 1960). Apparently, olfactory neurons undergo a more or less constant neurogenesis (Andres, 1969), whereby cells in different states of their lifespan co-exist (Farbman, 1992). Nevertheless, olfactory receptor cells can be relatively long-lived (Hinds, Hinds, & McNelly, 1984), underlining the complex structure of the olfactory system.

Increasing evidence shows that those odours which are not volatile are being processed by the accessory olfactory system in the vomeronasal organ before being transmitted to the accessory olfactory bulb (Ennis & Holy, 2015). Since studies in the framework of this thesis focussed on volatile odours, the system for processing non-volatile odours will not be explained any further. Still it is important to note the existence of an accessory olfactory system parallel to the main olfactory system.

A further unique quality of the olfactory receptor cells is their concomitant direct tangency of central nervous system and external environment due to lack of synaptic connection before projecting into the brain (Ding & Xie, 2015). Incoming olfactory information lacks thalamic gating and hence has direct access, amongst other core areas, to the cortical nucleus of the amygdala (Doty, 2001; Shepherd, 2007). Piriform cortex receives its main sensory input from mitral and tufted cells in the olfactory bulb instead of the thalamus (Wilson et al., 2015). Additionally, piriform cortex has three layers unlike cortices of the other sensory systems featuring six layers, and it is not topographically but highly associatively organised (Wilson et al., 2015). Apparently, activity within piriform cortex evoked by odours does not follow specific spatial patterns (Rennaker, Chen, Ruyle, Sloan, & Wilson, 2007; Stettler & Axel, 2009) as the case in the highly orientation-specific neurons of the visual system (*see for review*: Vaney, Sivyer, & Taylor, 2012). Olfactory processing in piriform cortex seems to be comparable to visual processing only in that sense that it has a parallel processing structure (Payton, Wilson, & Wesson, 2012). Hereby, single glomerulus activation is not sufficient for activity of neurons in piriform cortex; rather a threshold has to be reached by multi-activation of glomeruli (Davison & Ehlers, 2011).

1.4. Odours and memory

Early evidence of a special role for olfactory stimuli in memory comes from Proust (1922) describing a flashback into early childhood triggered by the smell and gustatory experience of a madeleine like his aunt used to bake (Proust, 1960). His description led to definition of the

'Proust phenomenon', lending the olfactory system significance concerning the potency of odours in triggering relatively old memory contents (Herz & Cupchik, 1992) as well as memories which are fairly emotional in nature (Adolph & Pause, 2012; Herz, 1998; Herz & Cupchik, 1995; Herz & Schooler, 2002; Willander & Larsson, 2007). Compared to visual and word cues, odours cue memories which are more closely related to the personal life and feature more affective associations (Hinton & Henley, 1993).

Neuronal correlates

As shown in section 1.3., the olfactory system has a direct reciprocal connection with several amygdaloidal structures. Since the amygdala gets rapidly activated by emotional, especially fear-related, stimuli and contributes to a large extent to memory consolidation of arousing and stressful events (Joëls et al., 2011; McGaugh & Roozendaal, 2002), its crucial involvement in the olfactory system's main circuits is conducive to the potency of odours to trigger affective responses and carve deep memory traces. Hereby, β-adrenergic activation of the receptors in the basolateral amygdala (lateral, basal and accessory basal nuclei) modulates influences of glucocorticoids and epinephrine, integrates information from the different structures and elements activated under stress (*see for review*: McGaugh, 2000) and mediates the amygdala's interaction with hippocampus (Strange & Dolan, 2004). Its crucial role within the circuit of olfactory stimuli processing makes odours prone to being well consolidated into long-term memory.

It could be shown that, in accordance with its significance in spatial memory, the hippocampus is involved in flexible orderly organisation of odours in a relational map (Bunsey & Eichenbaum, 1996; Dusek & Eichenbaum, 1997), which relates to its input from entorhinal cortex (EC). As a part of the memory system in the temporal lobe, EC has a crucial role in providing the hippocampal formation with information (Mayeaux & Johnston, 2004; Young, Otto, Fox, & Eichenbaum, 1997). Furthermore, EC exhibits evidence for internal memory structures (Young et al., 1997). As it receives direct input from mitral cells of the olfactory bulb (Doty, 2015) in combination with being a major input for hippocampal formation, its crucial role in odour memory is evident. Results of a study using field potential recordings in rodents propose that lateral EC modulates stimulus-specific, experience- and state-dependent olfactory coding (Xu & Wilson, 2012). This accounts for enhanced sensitivity to specific encoding under states of arousal and stress. Besides, EC is important for odour discrimination, mainly in dependency of location of novel and known olfactory stimuli, again underlining its relationship with the hippocampal system (Mayeaux & Johnston, 2004). Furthermore, the close connection of EC and hippocampal circuits is one of the

neurophysiological elements documenting a special role of olfactory stimuli in memory formation and consolidation.

1.5. Startle paradigm

Investigations involving stress and other affective states and judgements, e. g. of stimuli, often rely on explicit feedback and self-assessment of the participants in questionnaires. On the one hand this depends on the honesty of the participant when filling in the questionnaire beyond responding in favour of social desirability; on the other hand it involves a reflective self-monitoring process and the ability to correctly interpret one's own emotions, motives and characteristics. When stimuli like odours are involved, which may be processed subconsciously due to their lack of thalamic gating as described in section 1.3., it is of advantage to implicitly assess the affective state of a participant. This can be done for a person's judgement of a single stimulus as well as for the affective state of a person, for instance in a situation of stress and/or arousal, by means of the startle paradigm.

Our organism is equipped with reflexes which operate automatically, may not or only with great difficulty be repressed, and whose function is protection against potential threat and harm. One of these useful mechanisms is the startle reflex. In situations of abrupt and acute exposure to acoustic (loud noise), tactile (blow) or visual (flash) stimuli, the startle reflex helps us protect the organism against minacious injury (Koch, 1999). Even when the stimulus is acoustic in nature, the protection mechanism of the startle reaction involves an eye-blink response for eye protection due to cross-modal summation of acoustic, tactile and vestibular stimulation, accounting for a higher effectivity than proceeding within a single modality only (Yeomans, 2002). Furthermore, heart rate acceleration and flexor contraction of head – especially of dorsal neck muscles to prevent exposure of the dorsal surface (Yeomans & Frankland, 1996) – and skeletal muscles prepare for motoric action in terms of a "fight-or-flight" response (de Kloet et al., 2005), while sensory and cognitive processing and ongoing muscle activity come to an arrest (Graham, 1975). The origin of the startle response is not a zero baseline (Koch, 1999); condition, affective state, and external stimuli all contribute to the organism's responsivity. This is why even this most extensive reflex can easily be manipulated, that is enhanced (fear-potentiated startle; Hamm, Cuthbert, Globisch, & Vaitl, 1997; Lang, Bradley, & Cuthbert, 1990) as well as attenuated, by simple external and also internal stimuli (Koch, 1999). Since it is a protection mechanism, it can be enhanced by presenting aversive stimuli. Thus, negative pictures (Balada, Blanch, & Aluja, 2014; Bradley, Cuthbert, & Lang, 1993; Bradley, Codispoti, & Lang, 2006) or olfactory/chemosensory

stimuli (Pause, Adolph, Prehn-Kristensen, & Ferstl, 2009; Prehn, Ohrt, Sojka, Ferstl, & Pause, 2006) have the potency of pronouncing the startle response, whereas stimuli in these modalities eliciting positive affect can reduce it (*see for review*: Lang, Bradley, & Cuthbert, 1990; Schmid, Koch, & Schnitzler, 1995). Hence, the startle paradigm has become a valuable experimental tool for assessing sensor-motoric response patterns and internal affective states.

Fear-potentiated startle in experiments

In an experimental context, the startle response can be elicited by noise, air-puff on the eyes or electrical stimulation. By far the most commonly used startle setup in experiments featuring the greatest amount of data gathered so far (Koch, 1999) is the acoustic startle. The startle stimulus consists of a loud white noise with an instantaneous rise time. The startle response can be measured using electromyography (EMG), electrooculography as well as potentiometric, photoelectric or magnetic coils (Blumenthal et al., 2005). The most common method in psychophysiological research is measurement of action potentials on the surface via EMG. It is done with bipolar EMG compatible electrodes – typically sliver-silver chloride (Ag-AgCl) pelletised (Fridlund & Cacioppo, 1986) – attached to the lower orbital portion of the orbicularis oculi, a striated sphincter muscle surrounding the orbital fissure, of the left eye (Blumenthal et al., 2005). Preferential use of this surface method is owed to the sound correlation between its broad contraction detection and that of the muscle groups underlying the general startle response (Lawrence & De Luca, 1983). The EMG signal is conducted by skin, adding transduction liquid to the surface of the electrode before attaching it with adhesive collars (Fridlund & Cacioppo, 1986). The acoustic startle reflex has a very short onset of about 14-151 ms in humans (Yeomans & Frankland, 1996). The eye-blink reflex typically occurs at around 30-90 ms after startle stimulus onset, and its underlying mechanisms are thus considered pre-attentive. It has to be noted that startle responsivity can be influenced by a lot of different factors, amongst which are the intensity of the white noise used (Pilz, Caeser, & Ostwald, 1988) and the current motoric behaviour. Moreover it varies from one individual to another (Plappert, Pilz, & Schnitzler, 1993) with some individuals being startle stimulus resistant non-responders. Genetic prerequisites might partly be responsible for this (Paylor & Crawley, 1997).

Different models on pathways and areas involved in the startle response have been suggested, agreeing on a few basic elements. According to this, the neurophysiological basis for the startle reflex is a ponto-medullary pathway, from ventral cochlear nucleus through ventro-caudal pontine reticular formation to the spinal cord (Frankland, Scott, & Yeomans, 1995; Yeomans & Frankland, 1996). An indirect pathway from amygdala via rostro-lateral

midbrain to midmedulla apparently mediates the startle response (Yeomans & Pollard, 1993). Since the second pathway does not habituate, the startle response is most prone to emotional modulation when the first pathway is already habituating (Bradley, Lang, & Cuthbert, 1993). Due to its rapid action, the circuitry of shortest latency must be based on very few synaptic relays interconnecting cochlea and motor neurons, while rapid axonal transmissions must be responsible for signal transduction from cochlea to limb muscles (Yeomans & Frankland, 1996). These rapid interconnections account for the reflex-like appearance of the startle response, while involvement of the amygdala explains how modulation of this fast acting circuit by valent stimuli can proceed.

1.6. Aims of the dissertation

The experiments within the framework of this dissertation aimed at investigating different sensory variables in the influence of stress on memory enhancement. As stress is known to produce especially vivid and strong memories of stimuli central to the situation, fixation behaviour and its association with these processes were investigated (experiment 1). It was intended to shed light on whether stress causes different fixation patterns and hereby close the gap in finding a potential mediator in addition to cortisol between stress and memory. Despite its close association with amygdaloidal and memory structures, the potential of the olfactory system to create even stronger memory traces under stress has not been investigated yet. Hence, the potency of olfactory stimuli to produce emotional memory content which can be re-accessed pre-attentively with the startle paradigm was investigated as well as general startle responsivity one day after psychosocial stress (experiment 2). Finally, the findings concerning stress effects on odour memory and startle responsivity were extended to the acute stress phase in immediate aftermath of psychosocial stress (experiment 3).

2. Experiment 1: The influence of stress on eye fixation as a facilitator for memory enhancement

2.1. Introduction

The organism's stress response exerts an influence on memory enhancement not only in terms of affecting brain structures responsible for memory effects, but also with regard to manipulating attention prior to memory processes (Chajut & Algom, 2003). It was shown that stressed compared to control participants exhibit amplified visual area activation in response to pictures, reflecting higher-order visual processing (Heinze et al., 1994; Henckens, Hermans, Pu, Joels, & Fernandez, 2009; Moran & Desimone, 1985). Further evidence showed that very early indices of attentional processing (N1m and N1) are enhanced, as revealed in imaging studies (Elling et al., 2012; Shackman, Maxwell, McMenamin, Greischar, & Davidson, 2011), and that sensory input is potentiated during the state of hypervigilance caused by stress (Munk, Roelfsema, Konig, Engel, & Singer, 1996).

As described in section 1.2., memory is improved under stress, in particular for information relevant to the stressful situation, such as for the central objects used in the TSST (Christianson, 1992; Echterhoff & Wolf, 2012; Mather & Sutherland, 2011; Wiemers, Schoofs, & Wolf, 2012). While this facilitation of potentially meaningful information to get transferred into long-term memory has been concluded to be related to enhanced cortisol release (de Quervain, Aerni, Schelling, & Roozendaal, 2009; Lupien & McEwen, 1997; Quas, Yim, Edelstein, Cahill, & Rush, 2010; Roozendaal, McEwen, & Chattarji, 2009; van Ast et al., 2013; Wolf, 2009) attentional narrowing, directing focus towards salient items (Christianson, 1992), apparently also contributes to the memory effects. Several studies demonstrated stress leading to improvement of selective attention (Chajut & Algom, 2003) as well as attentional processes in connection with executive functioning in the brain (Beste, Yildiz, Meissner, & Wolf, 2013; Weerda, Muehlhan, Wolf, & Thiel, 2010). However, evidence for the opposite pattern was obtained in parallel (Arnsten, 2009; Plessow, Kiesel, & Kirschbaum, 2012; Shields, Sazma, & Yonelinas, 2016). These diverging results might have their origin in the underlying processes of the specific tasks used. While the results showing improvement are based on bottom-up processes, the opposite results were shown for top-down control based tasks. Stress induces domination of bottom-up, stimulus-specific attentional selection (Buschman & Miller, 2007) in order to focus on the most salient features and enhancing memory for these (Mather & Sutherland, 2011; Sutherland & Mather, 2012). In contrast, prefrontal top-down control is diminished (Arnsten, 2009; Sänger, Bechtold,

Schoofs, Blaszkewicz, & Wascher, 2014). This effect might explain the varied results. It is yet unclear whether these different forms of attentional processing are associated with different fixation patterns, such that stress exerts its influence not only internally, but also causes the external input to be processed in a modified manner due to focussing on other features, longer fixation times, and more frequent fixations.

Fixation is supposedly equivalent to orientation of attention during exploration of a scene (Henderson, 2007). When attention is directed towards the object fixated, its features are combined into a unified object representation (Treisman & Gormican, 1988). For forming a coherent object description, visual attention is a prerequisite (Rensink, 2000a, 2000b). Whereas object representations are retained in visual short-term memory – resistant to brief disruptions – unattended information is subject to rapid decay to be replaced by novel visual input, despite having been initially fixated (Rensink, 2000a). Subsequently, salient enough objects are potentially consolidated into long-term memory (McGaugh, 1966). These processes are thus based on fixation in concert with focal attention, both enhanced by stress-induced neurophysiological responses.

Experiment 1 aims at investigating whether psychosocial stress modifies fixation towards longer or more frequent fixation times, and if so, whether these measures facilitate subsequent memory processes assessed by free recall and object recognition tasks.

Hypotheses

Stressed participants are hypothesised to exhibit longer and more frequent fixation times on stimuli involved in the stressful situation, particularly for potentially relevant stimuli, that is central objects in the TSST. This, in turn, is presumed to be associated with enhanced memory performance in stressed compared to control participants and central compared to peripheral stimuli, on the next day.

2.2. Methods

2.2.1. Participants

The sample consisted of 63 non-smoking male (n = 32) and female students from the Ruhr-University Bochum, none of whom reported psychological or physiological diseases. Only women taking hormonal contraceptives (restricted to monophasic compounds with ethinylestradiol (0.02-0.035 mg) and gestagenic components) during their pill intake phase were included (Merz et al., 2012). The participants' age ranged from 18 to 34 years (M = 23.63, SD = 3.8) with a Body Mass Index (BMI) from 18.75 to 28.23 kg/m^2 (M = 22.93, SD = 2.53) due to cortisol related exclusion criteria, as mentioned in section 1.1.1. For participation

they were paid an expense allowance of 15 € or received course credits. The local ethic committee of the Faculty of Psychology granted approval for the study and the Declaration of Helsinki was followed.

2.2.2. Experimental procedure

After having signed informed consent, participants were randomly assigned to a stress or control condition. A modified version of the Trier Social Stress Test was applied for stress induction (TSST; Kirschbaum, Pirke, & Hellhammer, 1993; Wiemers et al., 2013). The control condition consisted of the friendly version of the TSST (f-TSST; Wiemers et al., 2012). Before stress induction, participants filled out the Positive and Negative Affect Scale (PANAS; Watson, Clark, & Tellegen, 1988) in a preparation room. A baseline saliva sample (-1 min) was collected by means of a Salivette® (Sarstedt, Nümbrecht, Germany). Participants were then led to a testing room where the experimenter fitted the eye tracking glasses to the participant and calibrated them. Until then, participants did not know which condition they had been assigned to. The experimenter left the room, which marked the onset of a 5 min preparation phase for either of the conditions. Upon their return after 15 minutes, the experimenter removed the eye tracker and led the participant back to the former room. Participants delivered the first post procedure saliva sample (+ 1 min) and again filled out the PANAS. During the final phase, participants engaged in a computer task extraneous to this investigation. At the end, the last saliva sample (+ 20 min) was collected and participants from the stress group were debriefed about the TSST being a standardised procedure.

On the second day, again the PANAS was filled out and a saliva sample to control for baseline cortisol concentration (d2a) was delivered. Afterwards, the experimenter asked participants to freely recall as many of the 20 objects present in the testing room. They were not informed about the number of objects, and after the free recall reported to have not been aware that their memory would be tested. Next, participants engaged in an object recognition task on the computer and delivered the final saliva sample (d2b). Finally, they were debriefed and remunerated (*Figure 2.1.*).

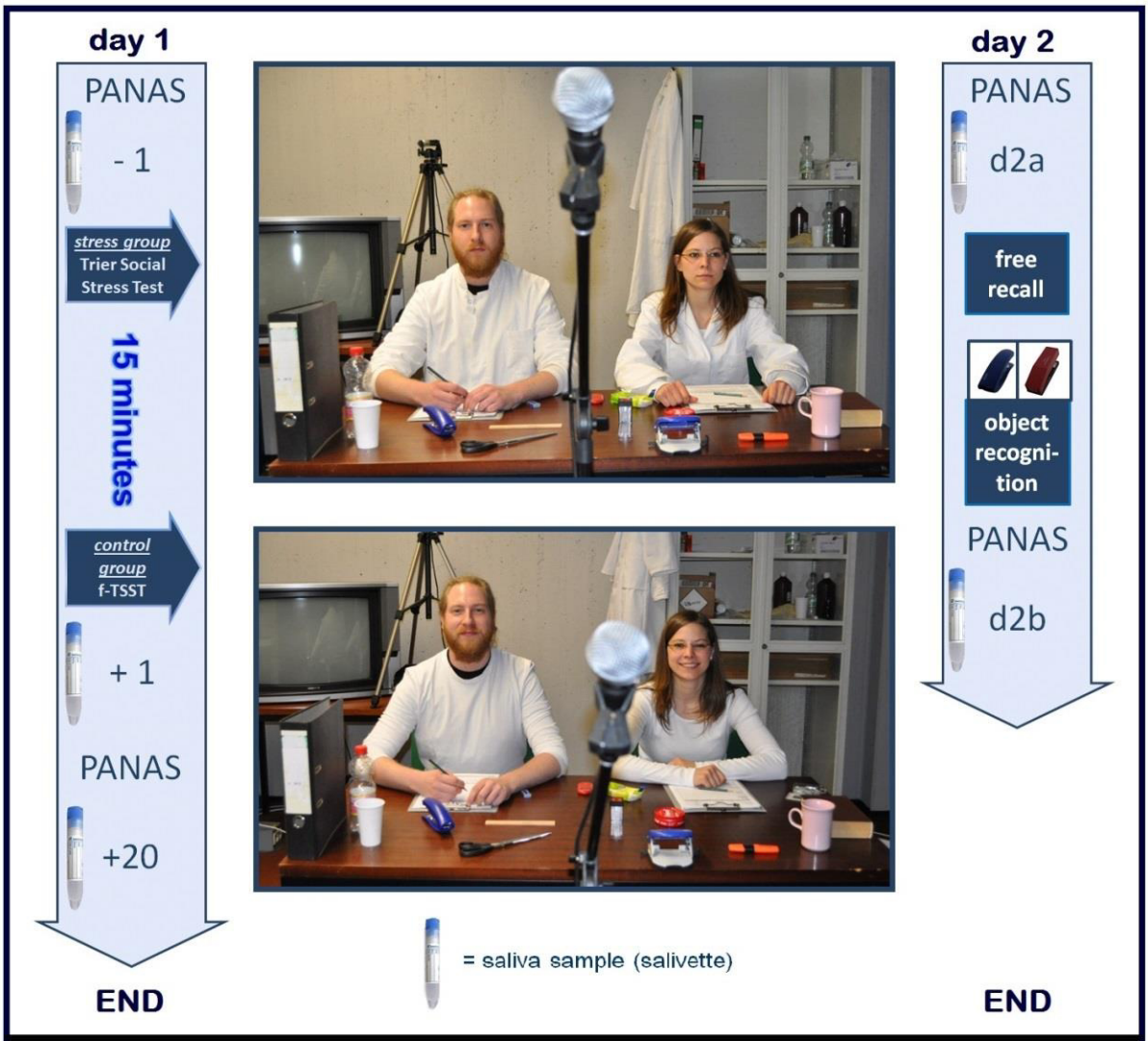

Figure 2.1. Experimental timeline and procedure of experiment 1.

2.2.3. Material

2.2.3.1. Stress and control procedure

Trier Social Stress Test (TSST)

Since the TSST (Kirschbaum et al., 1993) is the strongest of the established laboratory stressors (Skoluda et al., 2015), reliably activating SNS and HPA axis (Dickerson & Kemeny, 2004), as described in section 1.1.2., it was used for stress induction. In the slightly modified version used in all the here described studies, the TSST included objects which served as stimuli for memory assessment. Selected were 20 office items as they are most suitable to the laboratory situation, 10 of which were used by the committee members in a fixed time course, whereas 10 were present without being used. In order to avoid revealing the constructed and simulated character of the situation and in particular item usage, the speech part was extended

to 10 minutes (Wiemers et al., 2013; Wiemers & Wolf, 2015), such that objects would not be handled too often to make it obvious. Further, during arithmetic tasks, it was experienced that some people tend to close their eyes or gaze at the ceiling or wall in order to concentrate, which would have a mediating influence on fixation behaviour towards the objects and committee members. Hence, the arithmetic task was omitted in favour of a longer speech part.

On initiation of the preparation phase, participants were asked to fill in a self-estimation questionnaire on their general intelligence, presentation and maths skills, and individual appearance, on a 6-point scale. Subsequently, they took notes to prepare their free speech, but were not allowed to read those during the speech. Before the speech started, participants handed the self-estimation questionnaire over to the committee who used it to enhance psychosocial threat by studying it, taking notes on it, and revising the participants' rating during the course of the speech.

Friendly TSST (f-TSST)

The recently established f-TSST is a highly comparable control condition not activating the HPA axis (Wiemers et al., 2012). Previously used control conditions to the TSST mostly lacked comparability due to missing social interactions (e. g. Het, Rohleder, Schoofs, Kirschbaum, & Wolf, 2009) or dissimilar tasks and cognitive affordance. Similarly to the TSST, participants stand in front of a table, a male and a female person are seated at, featuring the same objects as in the stress condition. The male and female are introduced by their names and behave warm and friendly in contrast to the committee members of the TSST. Committee members are wearing white long-sleeved shirts for comparability with the stress condition and to avoid varying colours influencing the participant's attention or mood. In order to provide a similar preparation phase, participants are being advised to take notes for a general outline of the talk, while one of the friendly committee members leaves the room to avoid an awkward situation when the participant is finished taking notes or would like to start with their speech straight away. In this case, the other member remarks that the conversation can begin with the return of the colleague. Meanwhile, they engage in reading to maintain comparability to the stress condition that is not to engage the participant in a previous conversation and keep their attention off the objects on the desk. Instead of holding a monologue, participant and friendly committee members then engage in a mutual interaction. The participant can select a topic with suggestions being provided which resemble the content of a job interview such as curriculum vitae or career aspiration. The situation is not videotaped.

2.2.3.2. Physiological stress measures

Salivary cortisol

To assure a reliable measurement, participants were instructed in advance to their appointed time to refrain from taking any form of drugs or medication, as well as from drinking alcohol or engaging in excessive sports one day and from drinking anything except water and brushing their teeth one hour before the appointed testing time. Five saliva samples, three on the first and two on the second day, were collected using Salivettes®. Subsequently, samples were deep-frozen at -18 °C and analysed using a Dissociation-Enhanced Lanthanide Fluorescent Immunoassay (DELFIA) as described elsewhere (Dressendörfer, Kirschbaum, Rohde, Stahl, & Strasburger, 1992). The detection limit for salivary cortisol was set to 0.5 nmol/L. Intra- and inter-assay coefficients of variation were below 13%. As previously explained in section 1.1.1., cortisol is released in a pulsatile fashion, following a circadian rhythm. Hence, testing was restricted to a time window from 9:00 a.m. to 12:30 p.m.

Salivary α-amylase (sAA)

To assess sympathetic activity, the enzyme α-amylase (sAA) was analysed from the saliva samples (Rohleder & Nater, 2009). A colorimetric test with the substrate reagent 2-chloro-4-nitrophenyl-α-maltrotriosoide (CNP-G3) was applied for measuring sAA concentration (Lorentz, Gütschow, & Renner, 1999; Winn-deen, David, Sigier, & Chavez, 1988). Intra- and inter-assay variabilities were below 10%.

2.2.3.3. Affect measurement

Stress typically enhances negative subjective mood which can be assessed with questionnaires. The Positive and Negative Affect Scale (PANAS; Watson et al., 1988) includes 20 items, 10 of which express positive whereas 10 express negative affect. Participants rated the 20 emotional adjectives on a 5-point scale for their intensity, ranging from 1 = ‘very slightly or not at all’ to 5 = ‘extremely’. Answers result in a positive (PA) and a negative affect (NA) score. The German version of the PANAS exhibits a good up to excellent reliability quantified in a Cronbach’s α of .86 for the PA and .93 for the NA scale (Breyer & Bluemke, 2016). In the current sample, the internal consistency was good, marked by a Cronbach’s α of .834 for the PA and .806 for the NA scale.

2.2.4. Eye tracking recordings

The eye tracking recordings were done with SMI Eye Tracking Glasses 2.0 (SensoMotoric Instruments GmbH, Teltow, Germany) connected to the appendant notebook. The recording

screen was visible to the committee members for the whole procedure. The eye tracking device records the scene of the participant's field of view with an inbuilt camera. Two separate eye cameras on the inside of the frame record the participant's gaze according to the pupil position which is assessed by six infrared LEDs on each side. The scene camera has a resolution of 1280 x 960 pixels with a 60° horizontal and 46° vertical field of view. The gaze tracking range of the eye cameras is 80° horizontally and 60° vertically and their sampling rate was set to 60 Hz. Gaze position accuracy of the device is 0.5° across all distances. Since participants were standing at an approximate distance of 1 m, the distance from the participant's eyes to each of the AOIs has a variation of approximately 1.30 to 1.70 m. In consideration of the eye tracker's accuracy of 0.5°, the error margin is below 1.5 cm, resulting in sufficient accuracy to track even the smallest objects used in the study (e. g. the rubber). The eye tracker fuses the images of the three different cameras, such that the participant's fixations are mapped as focus circles onto the scene image. The glasses were adjusted for each participant individually with different variable nose pads and an adjustable bandeau. The participant was asked to focus on three target points positioned to cover distances and space of the field of view during TSST/f-TSST. The fixation point was displayed as a focus circle on the output screen of the eye tracking notebook where it was aligned with the actual locus the participant fixated on. This was repeated for the two other points for a precise 3-point calibration. The recordings started with initiation of the preparation phase, five minutes before TSST or f-TSST, respectively, and were stopped by the experimenter when re-entering the room after 15 minutes. Data were processed with the corresponding SMI software iView 3.5 and analysed with BeGaze 3.5.90, both included in the SMI Experiment Center 3.5.

2.2.5. Areas of Interest

Objects relevant for the analyses were defined by marking them in BeGaze as Areas of Interest (AOI) to determine the stimuli whereupon participants' fixations would be compared. They included 10 central and 10 peripheral objects and the faces of the committee members. Whereas the central objects were used by the committee members, the peripheral objects were static objects at fixed positions on the table (Wiemers et al., 2013). Central objects included a beaker, two clipboards, two pencils, a candy box, a rubber, a sharpener, a shelf, a stapler, a timer and a water bottle. Peripheral objects included a book, clips, a mug, a folder, a puncher, a dustbin, a ruler, scissors, a text marker and tissues. A semi-automatic detection mechanism calculated significant fixations according to an algorithm provided by the software BeGaze, which is named "event detection". An "event" includes multiple image frames for a less time

consuming analyses. Verification was done by two independent raters, confirming fixation position relative to the AOI by mouse click. The software calculated the total and average fixation time (in ms) for each AOI according to the rater's verifications. Adding up fixation duration on an item does not necessarily represent a better memory encoding of the object. Gain of information about the object could become saturated after having fixated it for a certain amount of time. Thus, the number of fixations on central and peripheral objects was also included in the analyses.

2.2.6. Memory assessment

On the next day, participants were asked to unexpectedly freely recall as many objects as they could remember having been present the day before in the testing room. Being common office items, their naming was unambiguous such that the experimenter simply checked the boxes beside the object names on a list when named by the participant. To make sure that any sort of memory task was indeed unexpected to the participant, they were asked whether they had expected a memory test. The number of items remembered was calculated separately for central and peripheral objects. Subsequently, participants underwent a computerised object recognition task. The 20 objects which had been in the room the day before, 20 similar objects differing in shape and colour, and 20 unrelated distractor objects were presented for two seconds each (Wiemers et al., 2013). Participants were instructed to indicate on a 6-point scale how sure they were to have seen the exact object, concerning shape as well as colour, the day before (1 = 'very sure to have seen the object'; 6 = 'very sure to have not seen the object'). Scores were summed up, again separately for central and peripheral items.

2.2.7. Statistical analyses

Mean values of all variables were calculated separately for stress and control group and, for the parameters concerned, separately for central and peripheral objects. In case of violation of the normal distribution, the data were log-transformed.

For comparison of memory performance between stress and control group, a discrimination index (DI) was calculated from the raw data of the object recognition task. Therefore, all replies indicating the respective object to have been present the day before were categorised as "seen", regardless of the level of certainty, and "unseen" in the opposite case (Green, & Swets, 1966/1974). Participants' correct categorisation of an item present during the TSST as remembered counted as a hit, whereas an item mistakenly categorised as remembered resulted in a false alarm. Hit and false alarm rates were calculated from these

variables in relation to the sum of actual and distractor objects. The DI consists of false alarm rate subtracted from hit rate, which was done separately for central and peripheral objects.

For correlation analyses differential measures were calculated for cortisol and negative affect (NA). The Area Under the Curve (AUC_i) was calculated representing cortisol increase as measured over time, from baseline to 20 minutes after termination of the stressor (Pruessner, Kirschbaum, Meinlschmid, & Hellhammer, 2003). For the NA, a delta value was calculated by subtracting the pre-assessment score from that of the post-assessment.

The data were analysed using SPSS 22.0.0 for Windows (IBM Corp., New York, USA). They were computed and entered into repeated-measures analyses of variance (ANOVA) with between-subjects factors CONDITION (stress, control) and SEX (male, female). For physiological and affective stress measures, the within-subjects factor TIME (baseline (-1 min), +1 min, +20 min or pre- and post-assessment, respectively) were included. Fixation and memory data included the within-subjects factor object CATEGORY (central, peripheral).

2.3. Results

2.3.1. Participants

Of the 63 participants tested, five did not show up on the second day. As the calibration of the eye tracking device was not successful for one participant, the recorded data could not reliably be analysed. Finally, two participants were excluded, one due to outliers in baseline cortisol (> 3 standard deviations (*SD*) from the mean) and one due to being a cortisol non-responder (delta-cortisol value negative > 1.5 *SD*). The final sample thus included 55 participants, 29 in the control and 26 in the stress group.

2.3.2. Stress induction

2.3.2.1. Physiological stress measures

Salivary cortisol

Since the data lacked normal distribution, analyses were conducted with log-transformed data. In the following, Greenhouse-Geisser corrected *p*-values (ε = .625) are reported due to violation of the assumption of sphericity ($\chi^2(2) = 44.909$, $p < .001$). The repeated-measures ANOVA resulted in a significant CONDITION x TIME interaction ($F(1.08,62.5) = 16.563$, $p < .001$), with an increase of cortisol in the stress and a decrease in the control group (*Figure 2.2.*). This shows that the stress induction was successful. Moreover, a significant TIME x SEX ($F(1.08,62.5) = 7.154$, $p = .006$) interaction was found due to a more pronounced cortisol increase in men, whereas in women only at time point +20 significant group

differences were shown. A post-hoc t-test for all participants revealed no significant differences between the groups in cortisol level at baseline, but at time points +1 min ($t(53) = -4.037$, $p < .001$) and +20 min ($t(53) = -5.133$, $p < .001$).

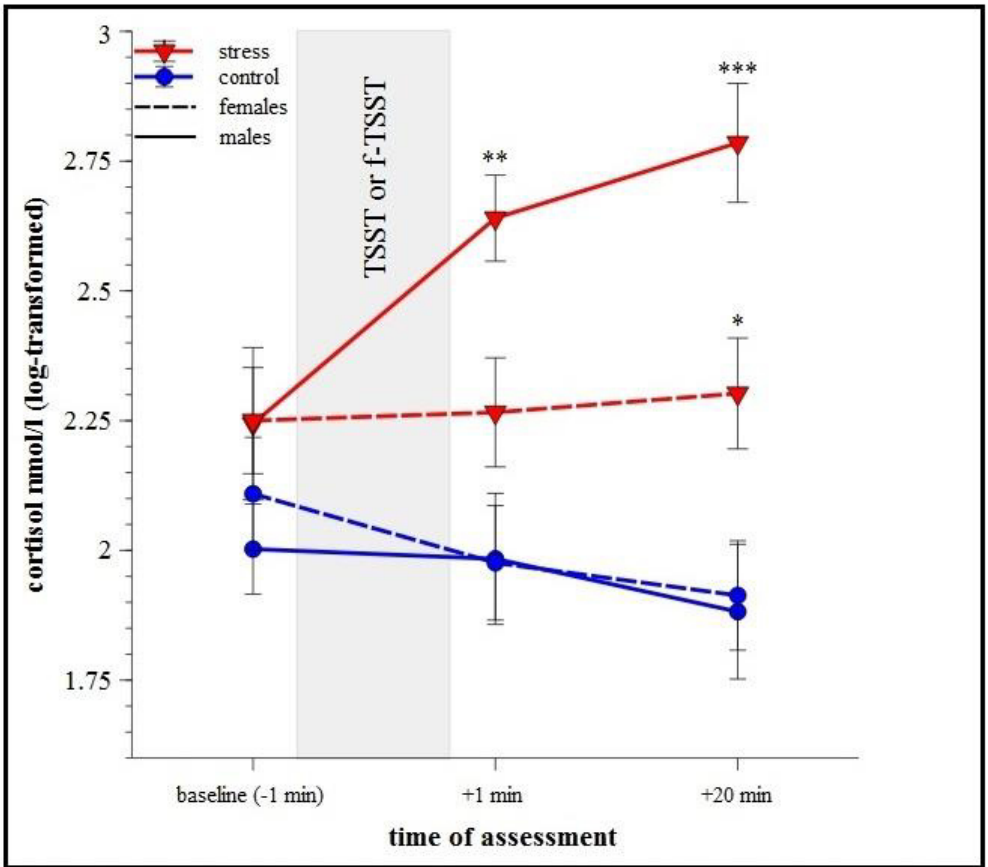

Figure 2.2. Time course of log-transformed cortisol data for both groups and sexes. Error bars refer to standard error of mean. Significances concern group differences. *$p < .05$ **$p < .01$, ***$p < .001$.

Salivary α-amylase (sAA)

The ANOVA for sAA showed a significant effect of TIME ($F(2,100) = 24.876$, $p < .001$), reflecting a significant increase in sAA release in both groups one minute, and a decrease 20 minutes after TSST or f-TSST, respectively (*Figure 2.3.)*. No other significant effects were detected.

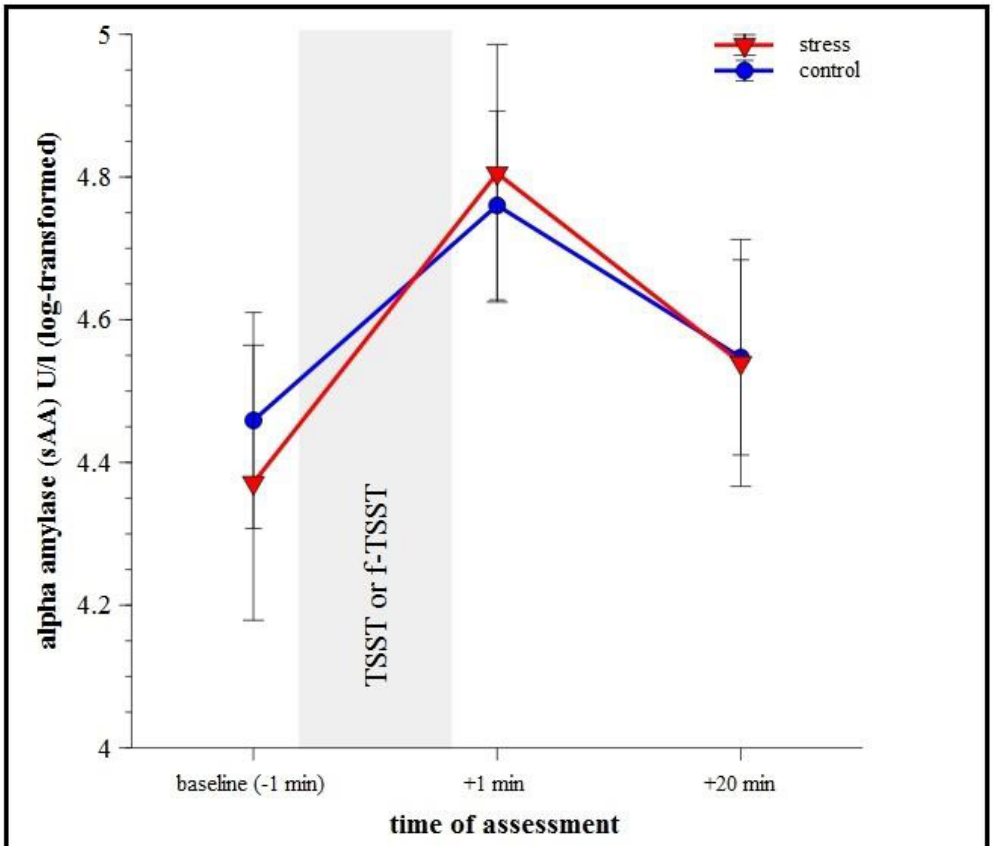

Figure 2.3. Time course of log-transformed sAA data for both groups. Error bars refer to standard error of mean.

2.3.2.2. Affect measurement

No differences in affect ratings for the pre-assessment were shown, but an increase in NA in the stress and a decrease in the control group for the post-assessment, whereas the PA showed no difference in the stress, but an increase in the control group (*Table 2.1.*). For the analyses of affect changes from pre to post-assessment with the PANAS, separate repeated-measures ANOVAs for NA and PA were performed with within-subjects factor TIME (pre, post) and between-subjects factors CONDITION (stress, control) and SEX (male, female).

Negative affect (NA)

For NA a significant CONDITION x TIME interaction was revealed ($F(1,51) = 21.408$, $p < .001$), showing an increase in NA from pre to post-assessment in the stress and a decrease in the control group. A post-hoc t-test showed that the groups did not differ in the pre-assessment ($t(53) = -.448$, $p = .656$), but significantly differed in the post-assessment of the NA ($t(53) = -5.028$, $p < .001$). Thus, negative affect was induced by the TSST.

Positive affect (PA)

For PA, the ANOVA resulted in a significant CONDITION x TIME interaction ($F(1,51) = 8.778$, $p = .005$) and a significant within-subjects effect of TIME ($F(1,51) = 10.427$, $p = .002$), with an increase in the control and no change in the stress group (*Table 2.1.*). In a post hoc t-test no differences between the groups were shown for the pre-assessment ($t(53) = .587$, $p = .559$), but significant group differences for the post-assessment of PA ($t(53) = 2.982$, $p = .004$). There were no sex effects in affect measures.

mean (SD)	stress	control
NA-pre	13.35 (3.21)	12.90 (4.12)
NA-post	16.58 (5.74)	10.72 (1.56)
PA-pre	29.50 (5.59)	30.34 (5.08)
PA-post	29.65 (6.39)	34.52 (5.71)

Table 2.1. Mean and standard deviation (SD) for negative affect (NA) and positive affect (PA) before (pre) and after (post) the experimental manipulation.

2.3.3. Memory performance

Free recall

For the free recall task a significant main effect of CONDITION ($F(1,51) = 48.740$, $p < .001$, $\eta^2 = .489$), with superior memory performance in the stress group, was revealed. The ANOVA further resulted in a significant within-subjects effect of CATEGORY ($F(1,51) = 115.810$, $p < .001$, $\eta^2 = .694$), with more central than peripheral objects recalled. Additionally, a significant CONDITION x CATEGORY interaction ($F(1,51) = 29.309$, $p < .001$, $\eta^2 = .365$) was found (*Figure 2.4.*). The stress effect on free recall memory performance was more pronounced for central than for peripheral objects.

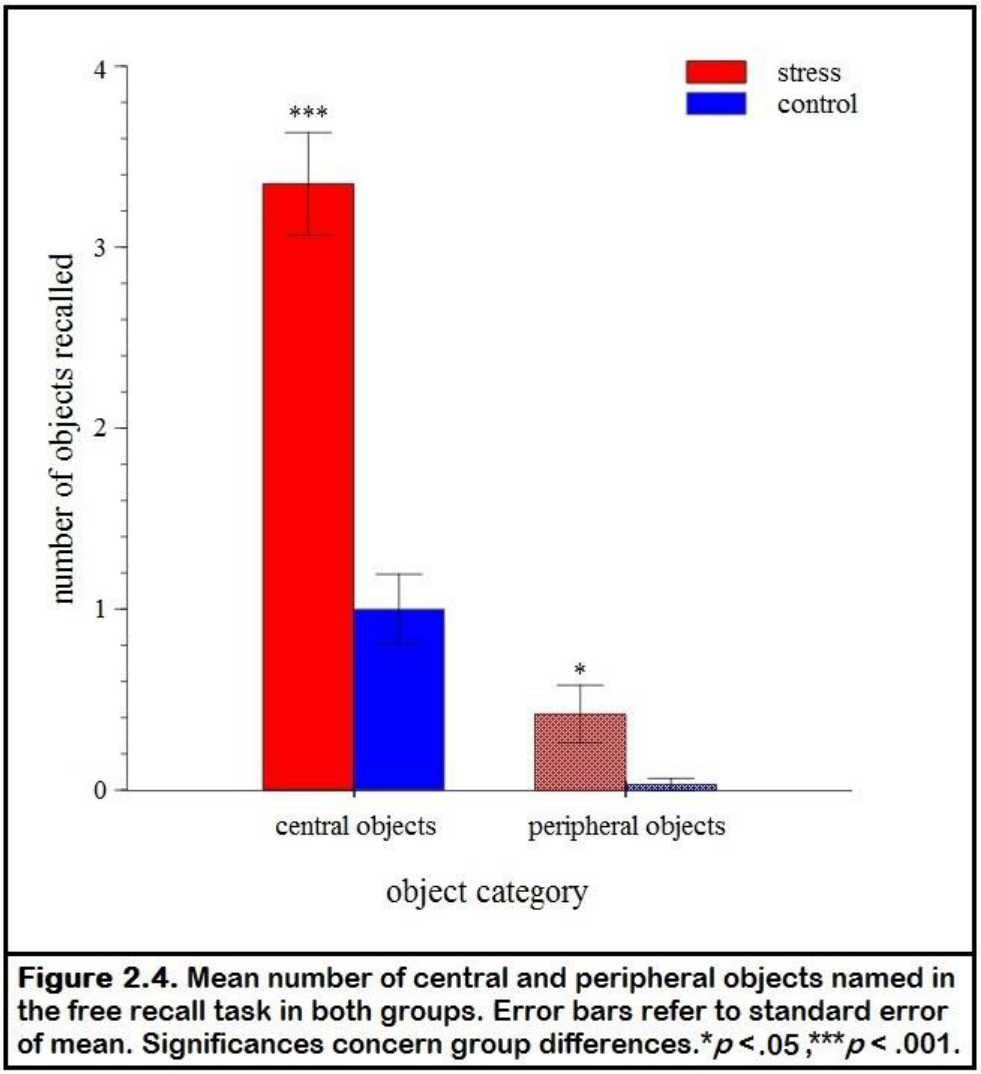

Figure 2.4. Mean number of central and peripheral objects named in the free recall task in both groups. Error bars refer to standard error of mean. Significances concern group differences.*$p < .05$;***$p < .001$.

Discrimination index (DI)

For memory, as assessed in the object recognition task, a main effect of CONDITION was shown ($F(1,51) = 6.321$, $p = .015$, $\eta^2 = .110$), with better recognition performance of stressed compared to control participants, for both central and peripheral objects (*Figure 2.5.*). Moreover, a significant within-subjects effect of CATEGORY was revealed, showing a better recognition memory for central than for peripheral objects in both groups ($F(1,51) = 75.041$, $p < .001$, $\eta^2 = .595$).

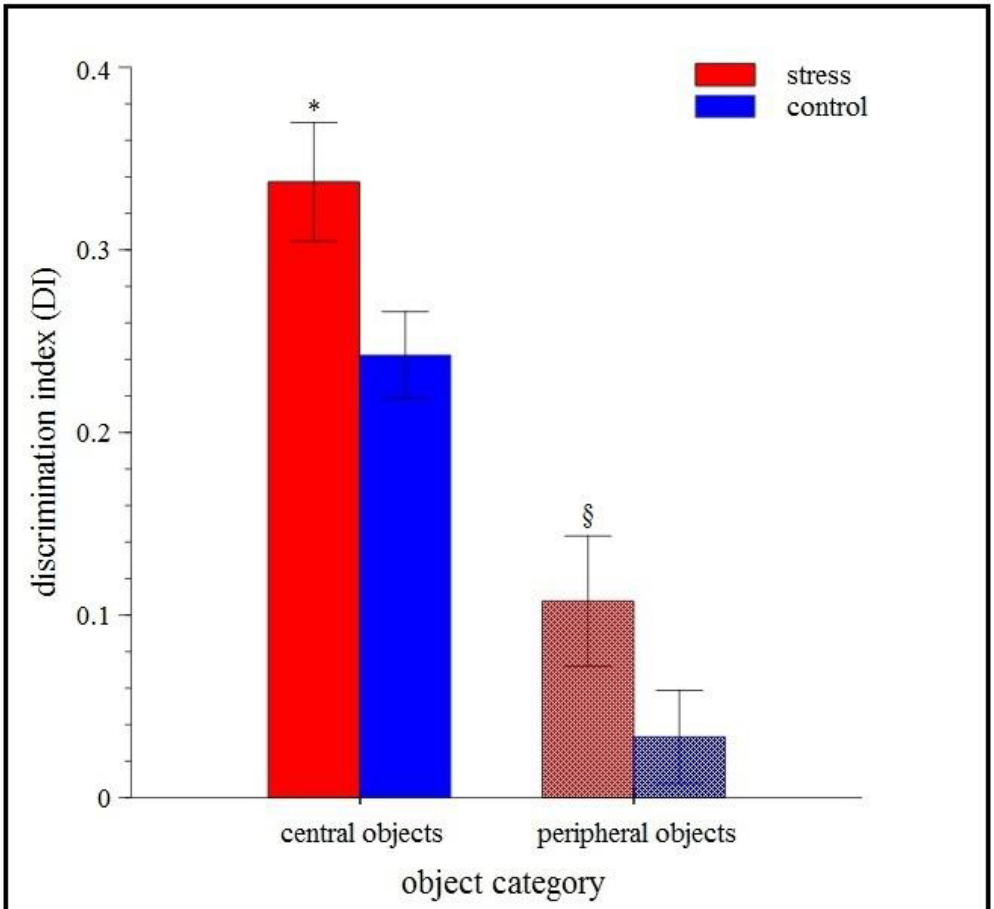

Figure 2.5. Discrimination Index (DI) for central and peripheral objects recognised by stress and control groups in the object recognition task. Error bars refer to standard error of mean. Significance concerns group differences.*$p < .05$, §: trend ($p = .091$).

2.3.4. Fixation measures

Areas of Interest

As the semi-automatic detection mechanism in BeGaze was operated by two different raters, an intraclass-correlation coefficient was calculated for fixation duration on the AOI (MacLennan, 1993). The correlation coefficient of $r = .995$ demonstrated excellent reliability of the raters' judgements about the fixation parameters.

Total fixation duration

The total fixation duration on the AOI was calculated separately for central and peripheral objects. A main effect of CONDITION ($F(1,51) = 15.324$, $p < .001$) showed longer total fixation times on both central and peripheral items by participants from the stress compared to the control group. The ANOVA further resulted in a significant main effect of CATEGORY ($F(1,51) = 39.603$, $p < .001$) and a significant CONDITION x CATEGORY interaction

($F(1,51) = 15.153$, $p < .001$). Fixations on central objects were longer than on peripheral objects, resulting in a significant stress effect on fixation duration only for central objects (*Figure 2.6.A*).

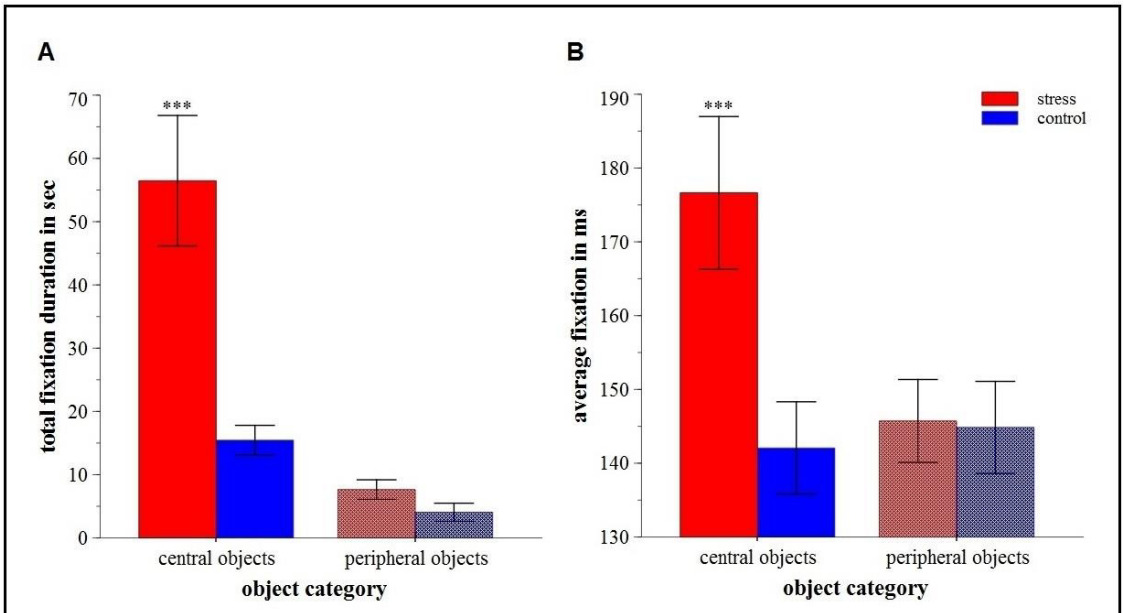

Figure 2.6. Mean total fixation duration in seconds (A) and average fixation duration in miliseconds (B) in stress and control group for central and peripheral objects. Error bars represent standard error of mean. Significances refer to group differences.* $p < .001$.**

Average fixation duration

For each AOI the average fixation duration was calculated and summed up separately for central and peripheral objects to be compared between the groups. The ANOVA revealed a main between-subjects effect of CONDITION ($F(1,49) = 4.194$, $p = .047$), again with longer fixation times in stressed than in control participants (*Figure 2.6.B*). Moreover, a main within-subjects effect of CATEGORY showed significantly longer average fixation duration on central than on peripheral objects ($F(1,49) = 8.400$, $p = .006$). A significant CONDITION x CATEGORY interaction ($F(1,49) = 8.743$, $p = .005$) reflected a significantly more pronounced effect in the stress compared to the control group.

Fixation count

The AOIs were fixated more often by stressed ($M = 164.5$, $SD = 140.12$) than control participants ($M = 60.4$, $SD = 62.98$). An ANOVA for number of fixations on central and peripheral objects showed a main effect of CONDITION ($F(1,51) = 14.443$, $p < .001$), with stressed participants fixating all objects more frequently than participants of the control group.

A within-subjects effect of CATEGORY ($F(1,51) = 53.257$, $p < .001$) and a CONDITION x CATEGORY interaction ($F(1,51) = 16.308$, $p < .001$) showed that this effect was more pronounced for central objects.

Central object fixation

As the categorisation in central and peripheral objects is based on manipulation by the committee members, the fixation behaviour during object manipulation was assessed. No significant group differences in object fixation by the time of manipulation were found ($F(1,50) = .013$, $p = .909$). Both groups fixated on average 7 of the 10 objects used by the committee members, [mean (SD)] 6.62 (2.86) in the control and 6.72 (2.48) in the stress group.

To verify categorisation based on object manipulation, fixation count and average fixation times before and from onset of object manipulation by the committee members were compared.[1] The results show that before the respective object was used, both groups exhibited less frequent fixations (stress: $M = 1.86$, $SD = 2.73$; control: $M = 1.34$, $SD = 3.54$) than afterwards ($M = 7.09$, $SD = 6.50$; $M = 3.54$, $SD = 8.46$). Average fixation times were also shorter before (stress: $M = 56.25$ ms, $SD = 55.00$ ms; control: $M = 36.68$ ms, $SD = 33.21$ ms) than after object use ($M = 127.75$ ms, $SD = 69.62$ ms; $M = 80.26$ ms, $SD = 46.16$ ms). A repeated-measures ANOVA with the factors TIME (pre, post) and CONDITION (stress, control) showed for fixation count a significant within-subjects effect of TIME ($F(1,51) = 30.681$, $p < .001$) and a significant CONDITION x TIME interaction ($F(1,51) = 5.144$, $p = .028$), demonstrating significantly more frequent fixations from the time of object manipulation than before, with a pronounced effect in the stress group. A similar significant within-subjects effect of TIME was revealed for average fixation times on the objects ($F(1,51) = 61.080$, $p < .001$). Moreover, a strong trend towards a CONDITION x TIME interaction ($F(1,51) = 3.787$, $p = .057$) was found, demonstrating significantly enhanced fixation times from the moment the objects became central compared to before, with a tendency of being more pronounced in the stress than in the control group. A main effect of CONDITION further reflected generally longer fixation times in stressed than in control participants ($F(1,51) = 7.755$, $p = .007$).

[1]Only 5 of the 10 central objects were considered for this calculation, since half of the items were central either from beginning of the trial (stop watch, clipboards, and pencils) or only at the very end (stapler, shelf). The remarkable differences of the mean to values across all objects are caused by excessive revisits of the clipboards the committee members were taking notes on, which are not included in this calculation. Further, objects not fixated were included, accounting for the low average fixation times.

2.3.5. Correlations between the variables

Partial correlations controlling for *condition* showed a strong correlation between fixation duration and number of fixations for central objects ($r = .934$, $p < .001$ for total and $r = .317$, $p = .020$ for average fixation duration), suggesting longer total fixation times to be owed to a more frequent fixation.

Despite longer average fixations and an increased memory performance in the stress compared to the control group, in particular for central objects, no bivariate correlation between fixation and memory was found. Furthermore, neither cortisol nor negative affect correlated with the two memory measures (*Table 2.2.*).

control variable: CONDITION		**cortisol (AUC_i)**	**NA_{delta}**	**average fixation**
free recall	*r*	-.108	.156	-.076
(central objects)	*p*	.440	.264	.585
DI	*r*	.016	.107	.107
(central objects)	*p*	.909	.444	.443

Table 2.2. Partial correlations of memory data for central objects with physiological (cortisol) AUC_i and affective (NA_{delta} = post- substracted from pre-assessment score) stress measures as well as average fixation duration for the central objects (*df* = 52).

2.3.6. Mediation analyses

In addition to the conducted analyses for correlations between the crucial variables, a formal mediation analysis was performed to investigate whether the influence of stress on memory was mediated by the average fixation duration. Since the strongest stress effects on memory were found for the free recall performance, a multiple regression analysis including the parameters stress, average fixation duration and free recall memory performance was conducted. No mediation was found (*Figure 2.7.*). The explained variation of the predictor *fixation* on memory had an estimate of $R^2 = 2.5\%$.

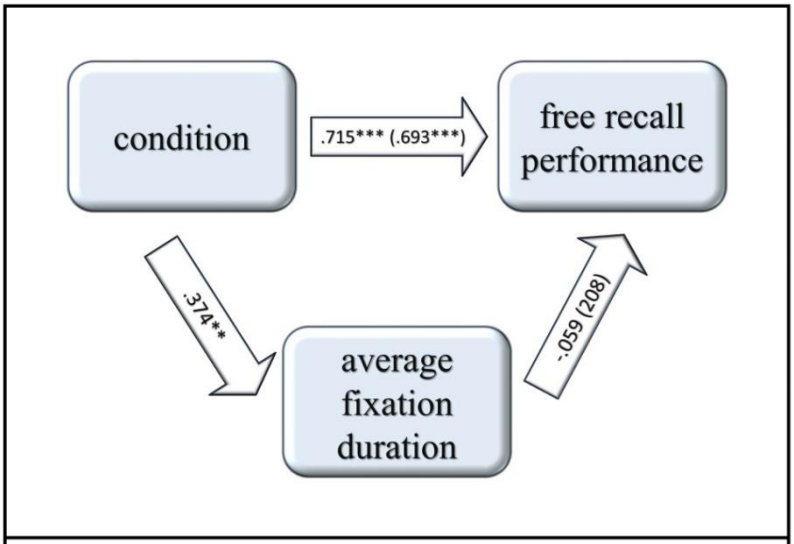

Figure 2.7. Mediation analyses with stress as independent variable, average fixation duration as the mediator, and free recall performance as the dependent variable. Path coefficients are the standardised *ß*-coefficients from multiple regression analyses, whereas the direct effects are represented by bivariate *ß*-coefficients in parentheses. $^{**}p < .01$, $^{***}p < .001$

2.3.7. Committee faces

The ANOVA for fixation duration on the committee faces showed a main effect of CONDITION ($F(1,51) = 4.641$, $p = .036$), with control participants exhibiting longer fixation times on the committee faces than stressed participants ($M = 65.20$ sec, $SD = 63.29$ versus $M = 36.03$ sec, $SD = 35.23$).

2.4. Discussion

The purpose of this study was to investigate the associations between stress effects on fixation behaviour and stress effects on memory, 24 hours after stress exposure. Both, fixation and memory measures were enhanced after stress in contrast to a non-stressful situation. However, there seems to be no direct relation between longer and more frequent fixations of the objects and better memory for them.

2.4.1. Stress induction

2.4.1.1. Physiological stress measures

Salivary cortisol

The data show an enhanced cortisol response after stress exposure in the stress group, but not in the control group. Before the experimental manipulation, the groups show no different

baseline cortisol. Thus, the stress induction was successful. Women exhibit a less pronounced cortisol response to the stress condition, but the female stress and control group differ significantly at time point +20 min. Their blunted cortisol response can be explained by their intake of oral contraceptives, as previously shown (Kirschbaum et al., 1999). Nevertheless, stress effects on memory in women were similar to those found in a previous study (Wiemers et al., 2013) and thus may occur even in the absence of a strong cortisol response. Additionally, there are no further variables co-varying with female cortisol measures, and no other sex differences were detected.

Salivary α-amylase

Both groups show comparable SNS activation as measured by sAA, which has been shown before (Wiemers et al., 2013, 2012). As both conditions include a social interaction, arousal is enhanced and thus is sAA release.

2.4.1.2. Affect measures

Stress effects are also due to the affective response (see sections 1.1.2., 1.2., and 2.2.2.3.), and affect ratings confirm a successful stress response in the stress group, in both males and females, with increase in NA in the stress group after the experimental manipulation and decrease of NA in the control group.

2.4.2. Memory measures

Memory performance in the stress group was superior to that in the control group, as previously shown (Wiemers et al., 2013). With the current investigation this could be demonstrated for recognition memory by using the object recognition task, as well as for recollection by means of the free recall task. The impact of stress was more pronounced with regard to the free recall task which is in line with previous findings of enhanced recollection in contrast to familiarity (Anderson, Wais, & Gabrieli, 2006; Kensinger & Corkin, 2003; Sharot, Delgado, & Phelps, 2004; Sharot & Yonelinas, 2008; Wiemers et al., 2013). Moreover, the stress effect was especially pronounced for central contrasting peripheral objects.

2.4.3. Object fixation

As hypothesised, stressed participants fixated the objects more often and longer than participants of the control group. Total, as well as average fixation times were prolonged in stressed participants, and fixation count showed a higher frequency. As the case for memory

measures, this effect was more pronounced and significant for central objects. Fixation times during object usage by the committee did not differ between the groups, thus, attentional focus on the items is comparable for the groups. However, from the moment they are used, particularly stressed participants exhibit even more pronounced fixation times on the objects than before they are used. This demonstrates the validity of the categorisation into central and peripheral objects. The differing fixation times after object manipulation suggest an association with attentional narrowing in stressed participants (see section 2.1.), as previously observed (Chajut & Algom, 2003). In contrast, more frequent fixations for emotional contrasting neutral and unusual slides have been observed before (Christianson, Loftus, Hoffman, & Loftus, 1991), but with shorter fixation times in the emotional condition than the two other conditions. This difference to the current study might be due to the objects used here being not innately emotional, but their context. In line with the current findings, though, fixation times were not correlated with memory outcome. According to another investigation, the number of fixations is a better predictor for memory outcome (hits) in contrast to fixation duration (Loftus, 1972). Interestingly, Loftus also found a covariation of memory performance with fixation duration, in line with the current findings.

2.4.4. Face fixation

People are experts in face detection and interpreting facial expressions. This makes faces highly salient emotional stimuli, rich in information (Bahrick & Lickliter, 2014; Caulfield, Ewing, Bank, & Rhodes, 2016; Crivelli, Jarillo, Russell, & Fernández-Dols, 2016). During a conversation as in the f-TSST, participants are thus expected to fixate the social agents often, i. a. for emotional feedback, whereas in the TSST participants are more likely to avoid eye contact. Since the committee members are the source of social evaluative threat and thus stress, due to their cold and neutral behaviour, they are perceived as aversive. Negative and threatening stimuli are mainly avoided (Wilson & MacLeod, 2003; Mogg & Bradley, 1998; Mogg, Mcnamara, Powys, Seiffer, & Bradley, 2000), as disengagement of attention is an essential self-regulation strategy (Posner & Rothbart, 2000). Additionally, a situation of social evaluative threat during a task that is prone to cause self-monitoring and motivation to perform well is most likely to elicit embarrassment and shame. In many different cultures gaze avoidance is an expression of this condition (Edelmann & Neto, 1989). This explains the dissociation of face fixation measures between the groups.

2.4.5. Relations between the variables

Partial correlations controlling for Condition showed no significant correlations of the stress markers (NA, cortisol, and sAA) or memory with fixation measures. Mediation analysis did not result in average fixation duration being a mediator between stress and free recall either. Despite the influence of stress on both, memory and fixation parameters, a powerful and direct relation between them could not be confirmed. Although only a few hundred milliseconds are needed to form lowest level representational structures of objects fixated (Rensink & Enns, 1998), without focussed attention, the object representations have an extremely restricted coherence in space and time (Rensink, 2000a, 2000b). Fixating an object thus does not guarantee the object representation to be consolidated when attention is not focussed towards it. The influence of stress on fixation patterns might be a prerequisite, but no warrant for object representations to be consolidated into long-term memory. There might be a direct influence on item information in visual short-term memory initial to the decision about their relevance for being consolidated into long-term memory. Experimental manipulation of fixations on stimuli is necessary to shed light on the causal relation between fixation behaviour and memory for these stimuli.

2.4.6. Conclusion

The study investigated effects of social evaluative threat on fixation measures and their association with memory outcome. The results confirmed the expected memory enhancement in stressed participants which was most prominent for central objects. As hypothesised, stressed compared to control participants indeed exhibited significantly longer total and average fixation times as well as a higher number of fixations on central objects. For face fixation, this pattern was reversed as an expression of gaze avoidance towards the committee members in stressed participants. Despite the influence of stress on both, memory and fixation measures, a direct relation between fixation and memory could not be confirmed. A stressful situation might create a vantage point in terms of modified fixation patterns associated with stress effects on memory at a later stage.

3. Experiment 2: Odour perception and memory 24 hours after its involvement in a stressful episode and enhancement of the startle response[2]

3.1. Introduction

Anyone has likely, for at least once in their lives, had the experience of a fulminating odour triggering very old and intense memories. Often, the memory for the odour itself is of a rather implicit nature such that it cannot be explicitly labelled despite the certainty of having been experienced before. As described in section 1.3., the olfactory pathway lacks thalamic gating (Doty, 2001; Shepherd, 2007). It is probably for this reason that odours can leave a strong implicit trace and in many cases cannot be specified, although well-known. This tendency of odours to manifest themselves without explicit awareness could be demonstrated in a laboratory setting applying the fear-potentiated startle paradigm. Samples of "anxiety sweat", collected from students before their oral examinations, and "exercise sweat", collected during ergometric training, were delivered via olfactometer in an auditory startle setup (Prehn et al., 2006). Even though only three participants were able to consciously distinguish the sweat samples from room air, startle responses were significantly enhanced for anxiety sweat in contrast to exercise sweat or room air. Thus, pre-attentive odour processing seems to have occurred accounting for a more pronounced startle reflex in response to the aversive odour (Prehn et al., 2006).

As described in section 1.3., odours have been found to trigger memories of emotional, personal, and mainly old autobiographical nature (Herz, 1998; Herz & Cupchik, 1992, 1995; Herz, Eliassen, Beland, & Souza, 2004; Herz & Schooler, 2002; Hinton & Henley, 1993; Willander & Larsson, 2007). Since the olfactory system maintains an intense connection to the amygdala which plays a prominent role in the effects of stress on memory, it seems likely that odours occurring within a stressful situation leave an exceptionally strong memory trace. In line with this, odours were proven to have the potential of triggering memory retrieval as contextual cues for long-term memory contents of a stressful situation (Wiemers, Sauvage, & Wolf, 2014). Apparently, odours could be associated with items experienced during the stressful episode, and re-experience of the odour reactivated the association facilitating access to long-term memory contents previously paired with it. In line with this, a functional

[2] The experiment has been published slightly modified as Herten, N., Otto, T., Adolph, D., Pause, B. M., Kumsta, R., & Wolf, O. T. (2016). Enhanced Startle Responsivity 24 Hours After Acute Stress Exposure. *Behavioral Neuroscience, 130*(5), 521-530.

magnetic resonance imaging (fMRI) study showed more pronounced activation in right anterior hippocampus, which is part of the extended olfactory network, for visual stimuli learned in combination with an odour compared to "visual-only" acquisition of these stimuli (Ghio, Schulze, Suchan, & Bellebaum, 2016). At a behavioural level, effects of that kind often remain undetected, particularly when odours are involved being hard to quantify. Alongside imaging methods, paradigms like the fear-potentiated startle described in section 1.5. offer the opportunity of assessing effects which might not be visible in behavioural measures.

The startle paradigm exhibits sensitivity to state of arousal (Balada et al., 2014) and to modification by stress. It was shown that during cold pressor stress the startle eye-blink response was similar compared to during baseline period, but diminished after stress induction (Deuter et al., 2012). Acute stress was argued to diminish the startle response in order to lend enhanced focussed attention to aspects of the stressful situation. In contrast, an inverted U-shaped pattern was suggested due to a low dose of cortisol (5 mg) enhancing and a high cortisol dose (20 mg) reducing the startle response in a pharmacological stress study (Buchanan, Brechtel, Sollers, & Lovallo, 2001). It is yet unclear whether emotional memory traces can be created by a one-time stress experience and one day later, when the physiological stress response has ceased, be triggered by re-experiencing stimuli present during the stressful episode on the previous day.

This study was designed to connect the enhancing effects of stress on memory with the potency of odours to trigger strong, often implicit, emotional memory traces. It aimed at demonstrating that an odour can be associated with a stressful situation and thus become an aversive stimulus, although previously unknown and neutral. As this effect might proceed implicitly, the startle paradigm, pre-attentively assessing affective states, was applied to detect emotional responsiveness to the respective odour. Moreover, effects of psychosocial stress on general startle responsivity one day after stress induction were investigated.

Hypotheses

It was hypothesised that 24 hours after stress induction enhanced startle responsivity in stressed compared to control participants can still be detected. Furthermore, the startle amplitude in response to the odour present during the stressful experience was predicted to be enhanced in the stress group. Additionally, participants stressed were assumed to show recognition memory for this odour superior to control participants, and further more aversive subjective ratings of it.

3.2. Methods

3.2.1. Participants

Participants included 70 non-smoking male (n = 36) and female students from the Ruhr-University Bochum. Exclusion criteria concerned mental and physiological diseases, smoking, regular medication use and intake of drugs exerting their influence via the HPA axis. Female participants were not tested during their time of menses or during pregnancy, and women taking hormonal contraceptives were excluded (Kirschbaum et al., 1999). The age of the sample ranged from 18 to 34 years (M = 24, SD = 3.65), with a BMI from 18.04 to 29 kg/m^2 (M = 22.65, SD = 2.74). Participants either received course credits or an expense allowance of 25 € for taking part in the study. The local Ethics Committee of the Faculty of Psychology approved of the study, and the Declaration of Helsinki was followed.

3.2.2. Experimental procedure

Participants were randomly assigned to either a TSST or f-TSST, without their knowledge about the condition. First, they signed informed consent and then filled in the Social Interaction Anxiety Scale (SIAS; Mattick & Clarke, 1998) in a preparation room. Afterwards, the first Salivette® (Sarstedt, Nümbrecht, Germany) was handed over for collecting the first saliva sample (-1 min), while participants filled out the PANAS (Watson et al., 1988). In the testing room participants were brought to, same sized towel strips had before been charged with a constant amount and concentration of an odour, methyl benzoate – in the following referred to as the target odour. The odour was dispersed using a ventilator the towel strips were attached to, while the participants held their speech (Wiemers et al., 2014). After the speech, participants were brought back to the preparation room where they rated their current affect using the PANAS and delivered another saliva sample (+1 min). Finally, participants filled in the NEO Five Factor Inventory (NEO-FFI; Costa & McCrae, 1992) and delivered another two saliva samples (+10 min, +30 min). Participants of the stress group were debriefed about the standardised procedure of the TSST, featuring fixed and constructed response patterns of the committee. At the beginning of the second day's testing session the PANAS was again filled in before initiation of the startle procedure. Participants pseudo-randomly started with either a visual or an olfactory stimulus presentation of the same temporal sequence and presentation times. Each block had approximately 17 min duration, during which the startle stimulus was applied on average 24 times in the presence and 42 times in the absence of a stimulus. Subsequently, participants rated seven odours for their pleasantness on a 4-point scale (1 = *very pleasant*, 2 = *pleasant*, 3 = *unpleasant*, 4 = *very*

unpleasant). Odours included a target odour and two distractor odours (section 3.2.7.1.) which had been delivered by means of the olfactometer for comparison of startle responses. Finally, participants were instructed to give a forced-choice reply on which of the seven odours they thought was the odour ambient in the testing room on the previous day (*Figure 3.1.*). The four other odours were included to mask the target odour and compare odour ratings. These were the unknown odour damascenone and the known odours lemon, lavender, and vanilla (Sulmont, Issanchou, & Köster, 2002).

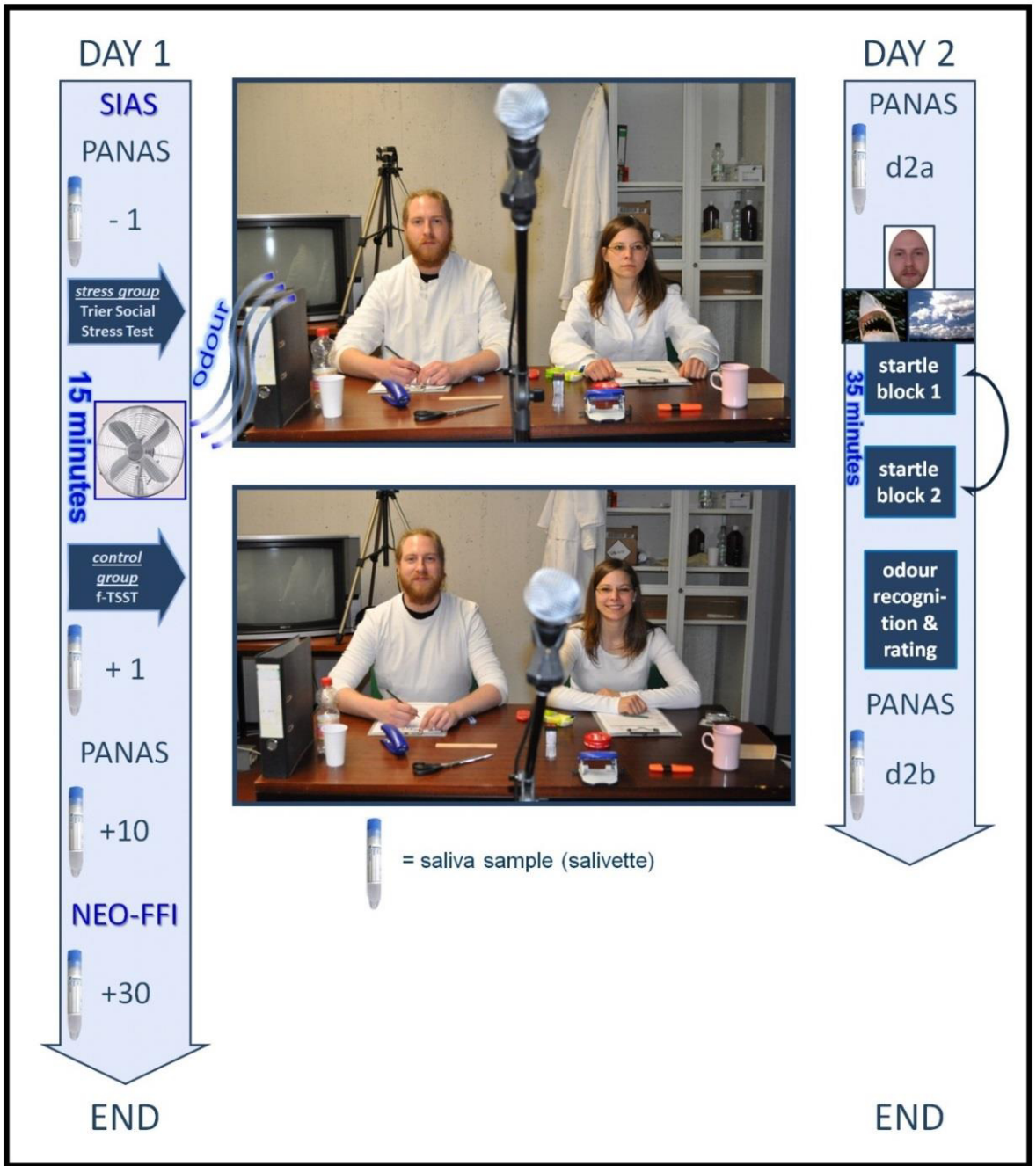

Figure 3.1. Experimental timeline and procedure of experiment 2.

3.2.3. Material

3.2.3.1. Stress and control procedure

Trier Social Stress Test (TSST)

For stress induction, the same modified TSST (Kirschbaum et al., 1993; Wiemers et al., 2013) as described in section 2.2.3.1. was used, including the same objects, material, and time course as in Experiment 1. For a detailed description of the procedure please see section 1.1.2. and 2.2.3.1.

Friendly TSST (f-TSST)

The control condition again consisted of the f-TSST (Wiemers et al., 2012) as described in section 2.2.3.1.

3.2.3.2. Physiological stress measures

Salivary cortisol

As described in section 2.2.3.2., participants received the instruction not to take any medication or other drugs, drink alcohol, or engage in excessive sports for 24 hr, as well as drinking anything except water and brushing their teeth 1 hr before testing. On the first day, four and on the second day two saliva samples were collected using Salivettes®. Subsequently, samples were deep-frozen at -18° C and analysed at our local biochemical laboratory using the *DEMEDITECs Cortisol Free in Saliva enzyme-linked immunosorbent assay (ELISA) Kit*, according to the manufacturer's manual. A coefficient of variation (CV%), expressed as the percentage deviation from the mean of ≤ 15% to retain any given duplicate sample, was used. Intra- and inter-assay CV were below 10%. As described in section 1.1.1., the release of the hormone follows a circadian rhythm. Thus, testing only took place in the afternoon, when the cortisol level has declined and stabilised, between 12:30 and 4:00 p.m.

Salivary α-amylase (sAA)

The sympathetic nervous system response was assessed with measuring the enzyme sAA (Rohleder & Nater, 2009; Rohleder, Nater, Wolf, Ehlert, & Kirschbaum, 2004). The measurement was based on an enzymatic action of the sAA using the substrate CNP-G3. At 405 nm the enzymatic action of sAA can be spectrophotometrically measured. With this method, sAA activity of the sample is directly proportional to the increase in absorbance at 405 nm. Intra- and inter-assay variabilities of the samples were both below 10%.

3.2.3.3. Affect measurement

Participants again provided ratings of their affect using the PANAS (Watson et al., 1988). For a detailed description of the questionnaire, please see section 2.2.3.3. As previously described, the ratings result in a positive (PA) and a negative affect (NA) score.

3.2.4. Trait questionnaires

Social Interaction Anxiety Scale (SIAS)

Since social interaction anxiety may have an influence on the stress response and might lead to stress responders in the control condition, participants were instructed to fill in the Social Interaction Anxiety Scale (SIAS; Mattick & Clarke, 1998) to control for this factor. The questionnaire includes 20 items to be rated on a 5-point scale for their expression, ranging from 1 = *not at all* to 5 = *extremely*. The German version of the SIAS exhibits a good reliability, quantified by a Cronbach's alpha of .90 (Heinrichs et al., 2002). In the current sample, the scale exhibited an excellent reliability with $\alpha = .962$.

NEO Five Factor Inventory (NEO-FFI)

The NEO Five Factor Inventory (NEO-FFI; Costa & McCrae, 1992) is based on a factor model (Borkenau & Ostendorf, 1985; Ostendorf & Angleitner, 1994) reflecting the most distinctive dimensions within the attribution of personality traits based on behavioural studies (Borkenau, 1988). The questionnaire assesses the value of the Big Five personality traits Neuroticism, Extraversion, Openness to Experience, Agreeableness, and Conscientiousness. The five scales consist of 12 items each, to be answered on a 5-point scale ranging from *strongly agree* to *strongly disagree*. The NEO-FFI was mainly applied to validate the answers given in the SIAS via the scale Agreeableness. The focus was further on the scale Neuroticism for comparing the groups on the basis of a reliable and significant personality parameter. The scales of the German version (Borkenau & Ostendorf, 1985) exhibit a good internal consistency with a Cronbach's alpha up to .85 (Neuroticism, Conscientiousness).

3.2.5. Startle evocation

A startle stimulus of 100 dB white noise and 50 ms duration with an instantaneous rise time was applied via 80 dB headphones (DT770M, beyerdynamic GmbH & Co. KG, Heilbronn, Germany). It occurred randomly in between 5 and 7 s after odour delivery or picture presentation onset, respectively. The white noise occurred in combination with a visual or olfactory stimulus at a 50% rate during trial and inter-stimulus intervals (ISI). The startle session consisted of two blocks, one with startle stimuli accompanied by pictures, the other by

odours. Participants randomly started with either the olfactory or the visual startle block to avoid habituation effects, even though the second startle response pathway usually does not habituate (Bradley et al., 1993), as described in section 1.5. To habituate the first response pathway for maximum emotional modification of the startle response via the second pathway (see above), both blocks started with a 30 s habituation phase including six startle stimuli.

3.2.6. Data recordings

Two bio potential electrodes (EASYCAP GmbH, Herrsching, Germany) were attached to the orbicularis oculi muscle of the left eye as described in section 1.5. for recording the startle amplitude (Blumenthal et al., 2005; Fridlund & Cacioppo, 1986; Lang et al., 1990). Additionally, a disposable ground electrode (GOLMED GmbH, Weddel, Germany) was attached to the forehead. The data acquisition device MP150 (BIOPAC Systems Inc., Essen, Germany) was used for transmission and amplification of the electrode signal, with filter settings of 10 to 500 Hz. The recording process was supported by the software MatLab (version R2012a, MathWorks Inc., Ismaning, Germany), providing the recording output files.

3.2.7. Startle stimuli

3.2.7.1. Olfactory stimuli

The odour essences methyl benzoate, bornyl acetate, and linalool (Sigma-Aldrich Co., Munich, Germany) were used as they had previously been rated as unfamiliar and neutral (Sulmont et al., 2002). Unfamiliar, as any experience with an odour can lead to an affective tone and thus influence rating and startle response to it; neutral, as the intention was to lend the odour an affective component through the stressful experience, which could only be tested if not already laden in the first place. The essences were dissolved in 50 ml scentless paraffinum liquidum, in concentrations of 60 µl for methyl benzoate, 850 µl for bornyl acetate, and 100 µl for linalool, in order to achieve comparable odour intensity (Wiemers et al., 2014). Odour delivery was done by means of an in-house-built 6-channel constant-flow (50 ml/s) olfactometer as described elsewhere (Lorig, Elmes, Zald, & Pardo, 1999) via oxygen masks covering nose and mouth. The mean latency of the olfactometer was measured as 447.5 ms for onset and 608.5 ms for offset of the odour essence in the mask. Activation of all channels was adjusted to the latency of the olfactometer to ensure maximum and constant intensity of odour delivery.

Since it had been demonstrated that participants comply well with breathing instructions and produce reliable respiration patterns (Adolph & Pause, 2012; Prehn et al., 2006), we

abstained from applying a breathing belt. Each odour was delivered for 7 s to provide sufficient dispersion time for inhalation (average respiratory frequency of an adult ≈ 12/min; Silverthorn, 2009). Additionally, a countdown was shown on the computer screen, counting backwards from 3 to 0 with the instruction to inhale when 0 is displayed. The respective odour channel was activated 500 ms before 0 was displayed to overcome the latency of the olfactometer's odour delivery into the oxygen mask. Each odour was presented seven times in a pseudo-randomised manner, such that no odour was presented twice in a row to avoid olfactory receptor habituation. The ISIs in between offset of the previous and onset of the next odour had durations of 20 seconds each. The countdown was randomly also shown in the ISI in order to prevent conditioning effects.

3.2.7.2. Visual Stimuli

As pictures have been proven to modify the startle response, a block with picture presentations was implemented for comparison. To directly compare startle responses in the two different modalities in relation to stress exposure, the picture stimuli included photographs of the committee members familiar to the participants from the previous day as well as unfamiliar committee members. These pictures were in-house made, showing the committee faces only, while other features like hair and background were masked. Since only two committee members were involved, each picture was repeated three times to ensure being provided with valid startle responses to both. Pictures of the International Affective Picture System (IAPS; Lang, Bradley, & Cuthbert, 2008) were included to control sensitivity and validity of the experimental startle setup in terms of fear-potentiated startle. Six positive (mainly landscapes) and six negative (mainly attacks) photographs were presented, which were matched for arousal within each category. These pictures were shown to have a robust effect on the fear-potentiated startle response in the previous study they were adapted from (Bradley, Codispoti, Sabatinelli, & Lang, 2001). Each of the pictures was presented twice, but only once combined with a startle stimulus to avoid any conditioning effects to the visual stimuli. The presentation screen had a measure of 15 inch x 12 inch with a brightness adjusted to 100 and a resolution of 1,280 x 1,024 pixels. In the ISI, a 20 x 20 pixels fixation cross was displayed at the centre of the screen. The participants were seated in a chair at an approximate distance of 45 cm from the screen.

3.2.8. Startle data processing

A semiautomatic mechanism of the software BrainVision Analyzer 2.0 (Brain Products GmbH, Gilching, Germany) was applied for selection of valid startle response data. The mechanism was set to detect startle responses occurring within a time frame of 50 ms to 225 ms from startle onset. The signal was baseline corrected (0–50 ms) and a 50 Hz notch filter was applied. Before applying a peak detection mechanism, the signal was rectified and finally manually verified. In total, 1.33% of the startle responses during odour or picture stimulus presentation were rejected due to reactions outside the expected time scope for startle responses (0.48%), artefacts of eye-blinks occurring close to the probable startle response (0–20 ms before or after startle probe onset; 0.43%), or non-responsiveness (amplitude did not exceed largest baseline amplitude by a factor of 2; 0.43%; Adolph & Pause, 2012).

3.2.9. Statistical analyses

For each single STIMULUS (three odours, four picture types) and each sensory MODALITY (olfactory, visual) mean values were calculated. A repeated-measures analysis of variance (ANOVA) with within-subjects factors MODALITY (odours, pictures) and between-subjects factors CONDITION (stress, control) and SEX (male, female) was conducted. For both modalities separately an ANOVA was repeated with ODOURS (3) x CONDITION (2) x SEX (2) or PICTURES (4) x CONDITION (2) x SEX (2), respectively. Statistical analyses were performed using SPSS 20.0.0.

3.3. Results

3.3.1. Participants

As the focus of this study lay on the impact of a stress-induced HPA response on startle responsivity, a stringent responder criterion was used which differed from the cortisol criteria used in experiment 1. A delta cut-off value was calculated by subtracting the baseline value from that of the saliva sample collected 10 minutes after termination of the stressor (+10), representing the peak of the hormonal response (Dickerson & Kemeny, 2004; Het, Schoofs, Rohleder, & Wolf, 2012; Kirschbaum & Hellhammer, 1994; Kirschbaum et al., 1993). For the current analysis, a stress response was defined as a difference score of $\geq$ 2.5 nmol/L, while a non-response was defined as < 2.5 nmol/L (Kirschbaum et al., 1992; Schoofs & Wolf, 2009; Wüst et al., 2000). This cut-off value caused exclusion of nine participants who were lacking a robust cortisol response in the stress condition and seven who expressed a cortisol increase to the control condition. Moreover, six participants were excluded due to being non-

responders to the startle stimulus, another seven participants due to technical issues with the electrodes, and one who did not show up on the second day.

The 40 participants remaining are equally distributed over the two groups. Since the menstrual cycle phase has an influence on the cortisol response as explained in section 1.1.1., women gave self-reports about the dates of their last three menses, such that the cycle phase at the time of the experiment could be calculated for group comparison. The 21 female participants included 7 who were in their luteal and 11 in their follicular phase, and 3 who were ovulating. To check for differences in the distribution of the different cycle phases in female participants between the two groups, the Chi-Square test was applied. No significant differences between the groups in menstrual cycle phase distribution were found ($\chi^2(2) = 1.25$, $p = .535$).

3.3.2. Physiological stress measures

Salivary cortisol

Due to a lack of normal distribution, the data were log-transformed. A repeated-measures ANOVA was conducted, with time of measurement as a within-subjects factor (baseline (-1), +1, +10, +30) and CONDITION (stress, control) and SEX (female, male) as between-subjects factors. Mauchly's test revealed a violation of sphericity ($\chi2(5) = 26.77$, $p < .001$), hence Greenhouse-Geisser corrected p-values ($\varepsilon = .654$) are reported. Cortisol responses show a successful stress induction, as participants stressed exhibit a rise in cortisol concentration, reflected in a significant CONDITION x TIME interaction effect ($F(1.96, 70.60) = 35.73$, $p < .001$), as well as a significant main effect of CONDITION ($F(1, 36) = 47.93$, $p < .001$) and TIME ($F(1.96, 70.60) = 12.89$, $p < .001$). Salivary cortisol levels at time points +1 ($t(38) = -6.588$), +10 ($t(38) = -8.181$), and +30 ($t(38) = -7.580$) showed significant differences between stress and control group (all $p < .001$), with a maximum difference at time point +10 (*Figure 3.2.*). Significant salivary cortisol baseline level differences between the stress and the control group were found ($t(38) = -3.083$, $p = .004$), which are accounted for by conducting an ANOVA with baseline cortisol response as a covariate proving them not to be the cause of any of the resulting group differences (see section 3.3.5.). Cortisol responses on the second day showed a significant within-subjects effect of TIME ($F(1,36) = 14.26$, $p = .001$) and a trend towards a CONDITION x TIME interaction ($F(1,36) = 3.33$, $p = .076$). Moreover, a main effect of CONDITION was detected ($F(1,36) = 10.35$, $p = .003$). Post-hoc t-test showed significant differences in salivary cortisol concentration between the groups before ($t(38) = -3.428$, $p = .001$) and a trend after testing ($t(38) = -1. 681$, $p = .101$).

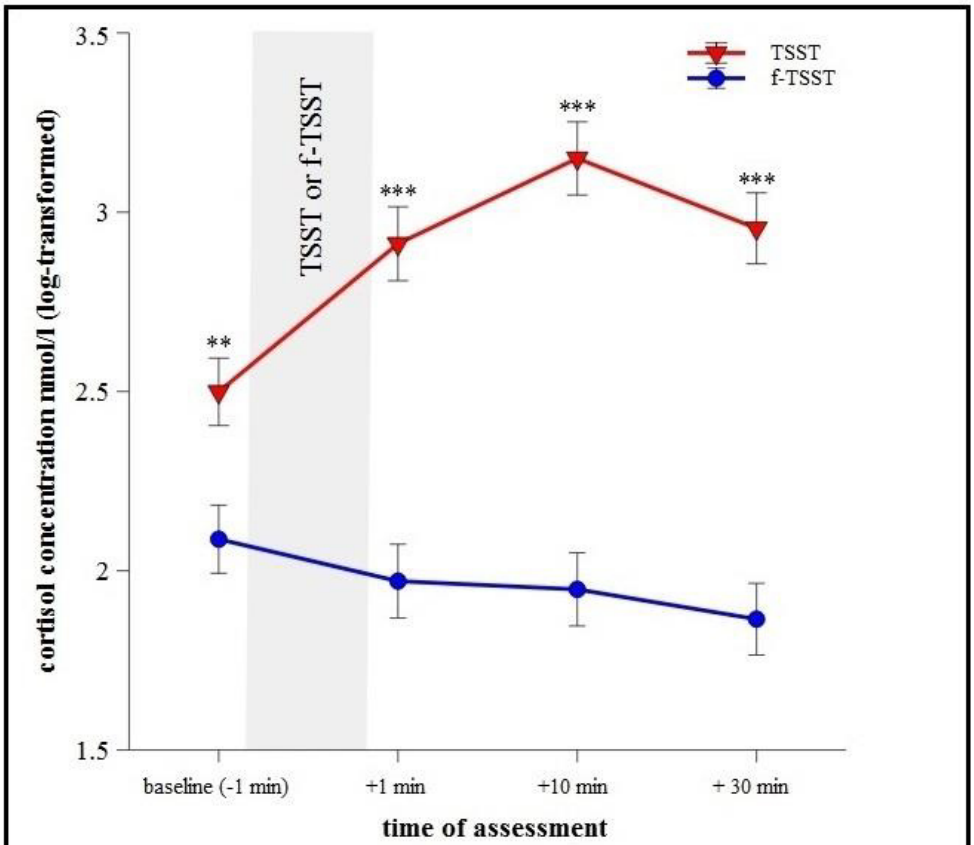

Figure 3.2. Time course of log-transformed cortisol data for both groups. Error bars refer to standard error of mean. Significances concern group differences.$^{**}p < .01, ^{***}p < .001$.

Salivary α-amylase

As the sAA data lacked normal distribution, they were log-transformed. A repeated-measures ANOVA was conducted with factors TIME (baseline (-1 min), +1 min, +10 min, +30 min) x CONDITION (stress, control) x SEX (male, female). A significant within-subjects effect of TIME ($F(3,108) = 22.5$, $p < .001$) for both groups was detected. Release of sAA peaked one minute after termination of the stressor, declining steadily (*Figure 3.3.*). Neither CONDITION ($F(1,36) = .075$, $p = .786$) nor the CONDITION x TIME interaction ($F(3, 108) = 1.765$, $p = .158$) showed significant differences, indicating a similar time course of sAA release for participants of both groups. No sex differences were detected.

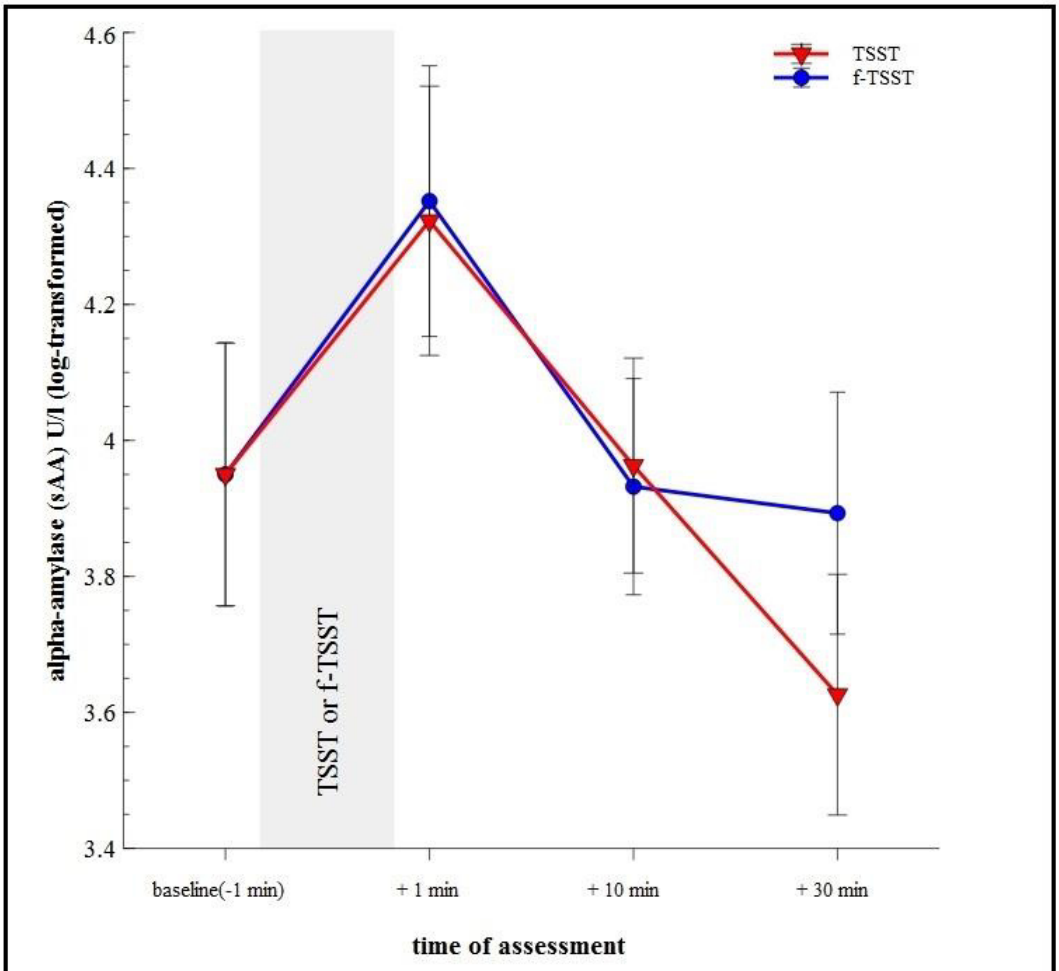

Figure 3.3. Time course of log-transformed sAA data for both groups. Error bars refer to standard error of mean.

3.3.3. Affect measurement

Data for pre- and post-assessment of the affect show no differences in affect ratings between the groups before the experimental manipulation. Participants of the stress group scored lower in PA and higher in NA than the control group after the experimental manipulation (*Table 3.1.*). A repeated-measures ANOVA with the within-subjects factors TIME (pre, post) and the between-subjects factors CONDITION (stress, control) and SEX (female, male) was performed separately for the two affect scales (PA, NA).

Negative affect (NA)

For the NA scale a significant two-way interaction of CONDITION x TIME ($F(1,36) = 9.63$, $p = .004$) was shown. A follow-up ANOVA comparing pre- and post-assessments of NA within the respective group resulted in a significant effect of TIME for both control ($F(1,18) = 9.53$, $p = .006$) and stress group ($F(1,18) = 4.48$, $p = .049$), with a decline of NA in the control and an increase in the stress group. There were no group differences in NA before the

experimental manipulation ($t(38) = .518$, $p = .607$). A follow-up comparison of pre- and post-assessments of NA between the groups revealed a significant difference with regard to post-assessment ($F(1,36) = 5.52$, $p = .024$). On the second day, stressed participants did not anymore show enhanced negative affect.

Positive affect (PA)

For PA, the ANOVA showed a significant within-subjects effect of TIME ($F(1,36) = 41.22$, $p < .001$), with both groups exhibiting a higher PA score after the experimental manipulation. There were no group differences in PA on the second day.

mean (SD)	stress	control
NA - pre	13.55 (3.25)	13.70 (2.77)
NA - post	15.10 (7.10)	12.25 (2.80)
PA - pre	27.45 (4.24)	31.00 (4.55)
PA - post	30.15 (9.60)	35.35 (5.72)

Table 3.1. Mean and standard deviation (SD) for negative affect (NA) and positive affect (PA) before (pre) and after (post) the experimental manipulation.

3.3.4. Trait Questionnaires

Social Interaction Anxiety Scale (SIAS)

No differences between the stress and control groups were revealed in social interaction anxiety as measured by the SIAS ($F(1,36) = .353$, $p = .556$).

NEO-Five Factor Inventory (NEO-FFI)

For the NEO-FFI, no differences in the measured factors Neuroticism, Extraversion, Openness to Experience, Agreeableness, and Conscientiousness between stress and control group were shown (all $p > .10$).

3.3.5. Startle responses

Since startle data were not normally distributed, they were log-transformed for the following analyses. A repeated measures ANOVA comparing startle responses between the two groups was conducted with between-subjects factor CONDITION (stress, control) and within-subjects factor STIMULUS (3 odours, 4 picture types). Mauchly's test resulted in a violation

of sphericity ($\chi^2(20) = 168.492$, $p < .001$), thus Greenhouse-Geisser corrected p-values ($\varepsilon = .302$) are reported.

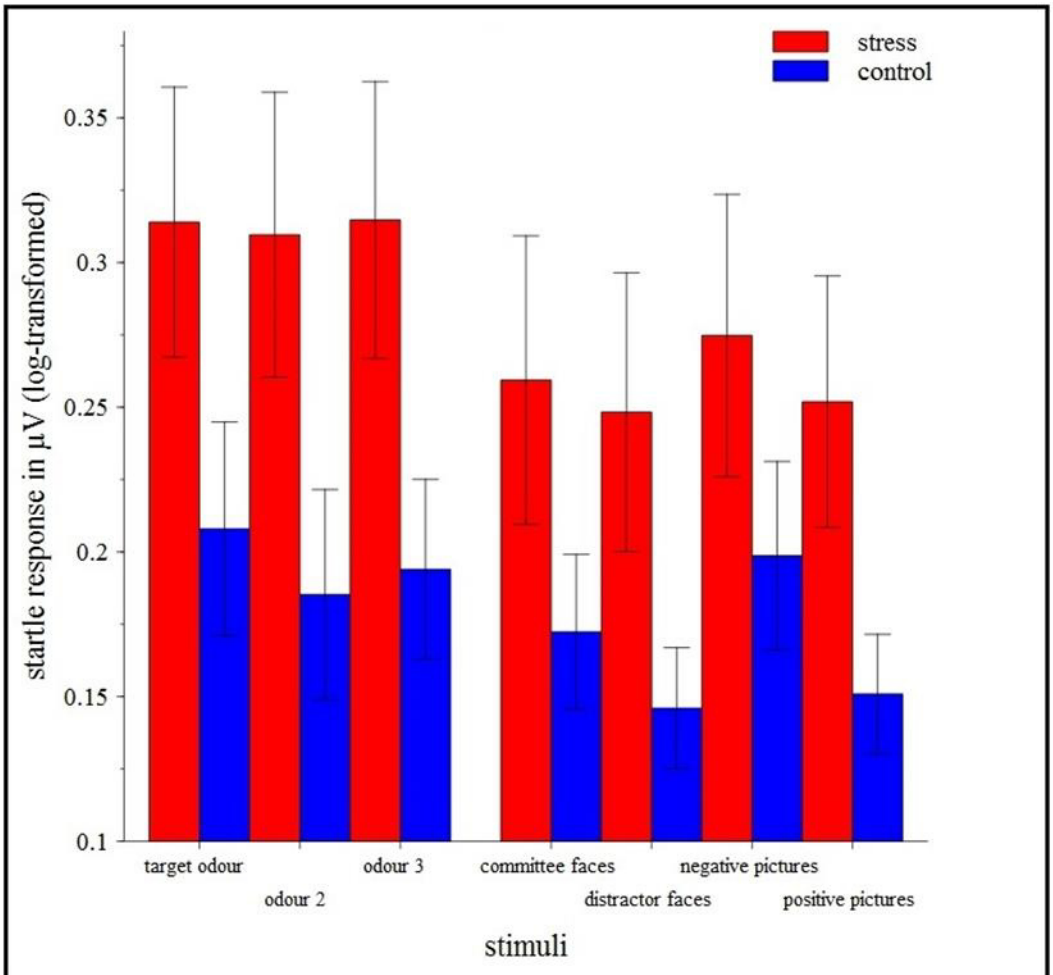

Figure 3.4. Mean log-transformed startle responses in both modalities (odours and pictures) for both groups. Error bars represent standard error of mean.
target odour = methyl benzoat, odour 2 = bornyl acetate, odour 3 = linalool

The ANOVA revealed a significant main effect of CONDITION ($F(1,36) = 4.530$, $p = .040$), indicating enhanced startle responsivity in the stress group across both modalities (*Figure 3.4.*). A trend for the within-subjects factor STIMULUS ($F(1.810,65.158) = 2.723$, $p = .078$) was detected. The ANOVA was repeated with baseline cortisol concentrations as a covariate, as described in section 3.3.2., due to cortisol baseline differences found between the groups. An even larger significance of the difference in startle responsivity between the groups when accounting for the baseline difference on the first ($F(1,35) = 8.730$, $p = .006$) as well as the second day ($F(1,35) = 9.199$, $p = .005$) could be shown.

3.3.5.1. Olfactory startle paradigm

A repeated measures ANOVA with the within-subjects factor ODOUR (target and two distractor odours) and between-subjects factors CONDITION (stress, control) and SEX (female, male) showed no differences in startle amplitude in response to the different odours ($F(2,72) = .326$, $p = .723$). The between-subjects factor CONDITION had a significant influence on the startle response ($F(1,36) = 4.339$, $p = .044$), but none of the other factors.

3.3.5.2. Visual startle paradigm

For pictures, the same ANOVA was conducted as for odours. It resulted in a main effect of PICTURE ($F(2.485,89.462) = 5.056$, $p = .005$), but only a trend towards a main effect of CONDITION ($F(1,36) = 3.160$, $p = .084$), and no CONDITION x PICTURE ($F(2.485,89.462) = .627$, $p = .570$), nor other interactions. A grouped comparison for picture types revealed a typical fear-potentiated startle; responsivity was significantly enhanced for negative compared to positive stimuli ($t(39) = 2.615$, $p = .013$). Moreover, startle responsivity to familiar committee members was higher than for unfamiliar committee members, and tended to become significant ($t(39) = 1.975$, $p = .055$).

3.3.6. Subjective odour ratings and odour recognition

In order to test for differences in affective quality of the target odour between the groups, the subjective ratings of the three odours from the participants of the two groups were compared conducting an ANOVA with CONDITION x odour RATING (3 odours). A trend towards a CONDITION x RATING effect was shown $F(2,68) = 3.071$, $p = .053$), as stressed rated the target odour more aversive than participants of the control group (*Table 3.2.*). Only participants of the stress group rated the target odour significantly more aversive than the two distractor odours, as revealed by a paired-samples *t*-test ($t(17) = 3.198$, $p = .005$; $t(19) = 6.469$, $p < .001$). Odour ratings of the distractor odours do not differ between the groups.

rating	condition	mean	SD
methyl benzoat	stress	3.10	.553
target odour	control	2.55	.826
bornyl acetat	stress	2.33	.594
distractor odour 1	control	2.55	.826
linalool	stress	1.75	.716
distractor odour 2	control	2.15	.745
damascenone	stress	2.40	.754
not included in startle block	control	2.50	.946

Table 3.2. Mean (SD = standard deviation) subjective odour ratings (1 = very pleasant, 2 = pleasant, 3 = unpleasant, 4 = very unpleasant) in both groups.

Testing for startle conditioning effects of odour delivery on odour ratings towards a more emphasised aversion, as found for heart rate data in a rodent study (Young & Leaton, 1994), ratings of each of the odours included in the startle block were compared to the rating of the neutral and unknown odour damascenone. This odour was encountered by the participants in recognition task and odour ratings, but not in the startle block. Data show that the differences in pleasantness ratings were not influenced by the odour having previously been paired with the aversive startle stimulus, for the ratings showed variation independently of that. Additionally, the rating of the target odour showed neither difference to the ratings of the startle distractor odours in the control group, nor to damascenone (all $p \leq .167$). Contrasting this, the target odour was rated significantly more aversive in the stress group than any of the other odours, independently of having been included in the startle block (all $p < .019$). Odour recognition performance showed no group differences; in each group the target odour was identified correctly as the odour ambient in the room on the previous day by only two participants. The remaining participants chose one of the other six odours included in the odour recognition task.

3.4. Discussion

This study aimed at investigating startle responsivity one day after exposure to social evaluative threat. General enhancing effects on responsivity as well as specific fear-potentiated startle in response to the odour present during the stressful episode were assumed. The data confirmed enhanced startle responsivity in stressed participants 24 hours after stress

exposure. However, response specificity to the odour was declined, reflected in similar responsivity to all odours delivered. As hypothesised, stressed participants rated the odour as more aversive than participants from the control group, but both groups showed an equally poor odour recognition performance.

3.4.1. Stress induction

3.4.1.1. Physiological stress measures

The stress induction was successful, reflected in cortisol rise only in stressed participants, peaking at +10 min as previously shown (section 1.1.2.), after application of a strict responder criterion with a cut-off value of 2.5 nmol/L to distinguish between stressed and non-stressed participants. The criterion led to exclusion of 20 participants, which is comparable to other studies using the TSST (e. g. Kirschbaum et al., 1993), even though slightly higher than in a previous study from the department (Wiemers et al., 2012). Accounting for baseline cortisol differences between the groups, an ANCOVA proved them not to be the cause of differences in startle responsivity. Further, there were no group differences in relevant personality traits, such as social interaction anxiety or neuroticism, as well as in sAA response (see section 3.3.4.), demonstrating their high comparability. Since metyrapone, repressing endogenous cortisol production, was shown to significantly increase startle responsivity (Roemer, Nees, Richter, Blumenthal, & Schächinger, 2009), cortisol might not mediate the stress effect on startle eye-blink at all. As suggested by the above mentioned authors, it might have been corticotropin-releasing hormone to have influenced the effect of metyrapone on startle responsivity, which might account for the lack of differences in startle amplitude between the groups despite cortisol baseline differences. It has to be acknowledged that HPA activation measured via cortisol is only one out of multiple factors composing the stress response.

3.4.1.2. Affect measures

Participants' affect ratings additionally confirm that stress successfully induced mood changes causing differences between stress and control group. Higher PA in the stress group after the experimental manipulation can be explained by included adjectives like "attentive" and "awake" which might be increased after the TSST, despite being part of the PA scale.

3.4.2. Startle responsivity

The findings of generally enhanced startle responsivity across all stimuli and both modalities extend the result of a previous study showing diminished startle eye-blink responses 45

minutes after stress exposure (Deuter et al., 2012) to a period of 24 hours. The current study shows that stress can have long-term effects on startle responsivity, even when the acute stress situation has terminated. In line with this, a one-time stress experience was demonstrated to have an effect on eye-blink conditioning 24 hours later in male rats (Beylin & Shors, 1998). Thus, emotional modulation by stress is not only apparent in immediate responding, but can be maintained over a 24 hour time-course. It was shown that the lateral/basolateral nucleus of the amygdala is involved in facilitating eye-blink conditioning during stress exposure, mediated via *N*-methyl-D-aspartate receptor activation (Shors & Mathew, 1998). Moreover, another rodent study showed a gradual build-up of avoidance behaviour up to 10 days after acute immobilisation stress accompanied by increased spinogenesis in the basolateral amygdala. These findings indicate that the effects on the startle response detected in the current study might be based on altered processing of the amygdala.

In contrast to the affective ratings, which did not differ between the groups on the second day, cortisol differences were observed. The ANCOVA, as described in section 3.3.2., showed that these differences did not contribute to the effects on startle responsivity in stressed participants. A context-effect due to the same preparation room and experimenter might be responsible for the cortisol response on the second day in stressed participants. As discussed on the basis of the results of the study of Roemer et al. (2009) in section 3.4.1.1., combined with the results of the ANCOVA, context-induced cortisol enhancement is very unlikely to have caused stress effects on the startle eye-blink response. The higher cortisol level in participants of the stress group on the second day was not reflected in the affect ratings, as previously mentioned. Thus, the lasting stress effect on the startle response and physiological markers occurred without conscious mood alteration. In line with this, it was previously reported that neuroendocrine factors might modulate the startle response without any subjective fear- and anxiety-related affect (Miller, McKinney, Kanter, Korte, & Lovallo, 2011).

The hypothesis that stressed participants would exhibit an even more pronounced startle response to the target odour methyl benzoate could not be confirmed. Rather, participants from the stress group seemed generally sensitised. Since the startle paradigm is very sensitive to detecting arousal and negative affect (Balada et al., 2014), the long-term effects of the stressor might have caused a “spill-over” effect, leading to a responsivity so strong that differential effects in response to the odours could not be detected. The low specificity in the stress group might have been caused by a shift of amygdala function shown in an fMRI study featuring highly negative movie clips (van Marle, Hermans, Qin, & Fernández, 2009). The

heightened sensitivity to potential threat shown in this study was accompanied by lower levels of specificity. The direct access of the olfactory sensory system to the amygdala might have caused an even more emphasised impact of its shift of function than the case for pictures, such that the difference in startle responsivity to negative and positive pictures was less affected. This would explain why startle responses to pictures might show a more distinctive specificity in comparison with responses to odours in the stress group. Descriptively startle responses to the pictures indeed are less specific in the stress than in the control group. This also becomes evident in the stronger startle response to the faces of the committee members known compared to the unknown faces in the control group, which might reflect a recognition effect as previously shown for skin conductance response and heart rate (Stormark, 2004). Furthermore, startle amplitudes in response to odours were generally more pronounced than in response to pictures which would also suggest a greater impact of olfactory stimuli in general. Since participants were wearing an oxygen mask for odour delivery, it cannot be ruled out that this might have caused an uncomfortable feeling contributing to more pronounced startle responsivity to odours than to pictures (Adolph & Pause, 2012). Nevertheless, the fear-potentiated startle response was successfully replicated from previous studies (Ameli, Ip, & Grillon, 2001; Bradley et al., 1993; Greenwald, Bradley, Cuthbert, & Lang, 1998; Paschall & Davis, 2002), indicating high sensitivity and validity of the experimental setup.

3.4.3. Subjective odour ratings and odour recognition

The hypothesis of a more aversive subjective rating of the target odour in participants of the stress compared to those in the control group could be confirmed at a trend level. Apparently, the target odour was negatively associated with the stress experience, indicating that the paradigm was successful. However, no memory differences were found in recognition of the target odour - only two participants in each group correctly identified the target odour. It seems to be an implicit effect the increased negative odour rating in stressed participants is based on.

3.4.4. Conclusion

The present study provides evidence for enhancement of human startle responsivity one day after exposure to an acute psychosocial stressor. This effect was found to be based on an overarching sensitivity at the expense of specificity for olfactory or visual stimuli experienced during the stressor, apparently associated with amygdalar responding in the aftermath of stress.

4. Experiment 3: Odour and object memory in the immediate aftermath of a stressful episode and its enhancement of startle responsivity

4.1. Introduction

A one-time stressful experience has the potential of generally enhancing the human startle response one day later, as shown in experiment 2 (Herten et al., 2016). However, through influence of amygdaloidal processes, specificity of the startle response declines – an odour ambient during the stressful episode in the TSST does not enhance the startle response on the next day. It was previously shown that acute stress can lead to a diminished startle response immediately after stress exposure (Deuter et al., 2012). Since the results from experiment 2 showed enhanced startle responsivity being accompanied by lowered specificity (Herten et al., 2016), more blunted startle responsivity might go along with enhanced specificity. Thus, the current experiment focusses on startle responsivity in the immediate aftermath of the TSST, expecting enhanced specificity to the odour present during the stressful experience.

In experiment 2, a more aversive subjective rating of the odour was shown, reflecting a long-term memory trace despite the lack of conscious recognition, which was poor (section 3.3.6.). In accordance with a possible reversion of effects towards a more pronounced specificity, odour memory in closer temporal proximity to its experience is hypothesised to be improved in comparison to 24 hours later and better in the stress than in the control group.

Experiment 1, as well as previous studies with the same setup (Wiemers et al., 2013; Wiemers & Wolf, 2015), have shown that memory for central objects of a stressful experience is enhanced 24 hours later in comparison to a control situation. Despite the increase of fixations and fixation duration in the stress group in experiment 1, the data did not correlate with memory outcome, nor did they mediate the relation between stress and memory. Apparently, memory of a stressful episode becomes increasingly consolidated over a time-span of 24 hours. There is not much known yet about stress effects on memory for objects involved in the stressful situation in the immediate aftermath of the stressor, before consolidation processes come into action. Rapid effects of stress on vigilance and attention as described in the introduction (section 1.1.) are suggestive of beneficial effects on shortly delayed free recall and recognition memory. Experiment 3 was conducted to assess stress effects on memory in the acute phase in contrast to those one day later, in combination with stress effects on immediate startle responsivity.

Hypotheses

It is hypothesised that participants of a psychosocial stressor in its immediate aftermath exhibit enhanced startle responsivity to an odour ambient during the stressful experience, reflecting more pronounced specificity than 24 hours after the stressor. Memory for the odour as well as for central objects of the stressful situation is hypothesised to be enhanced due to increased vigilance caused by acute stress.

4.2. Methods

4.2.1. Participants

Participants included 70 non-smoking male (n = 35) and female students from the Ruhr-University Bochum. Exclusion criteria were mental and physiological diseases and regular medication use, a BMI under 18 or over 29 kg/m^2. Only free cycling women having a regular menstrual cycle were tested, outside menses and pregnancy.

The participants' age ranged from 18 to 33 years (M = 23.59, SD = 3.62) and their BMI from 18.07 to 28.99 kg/m^2 (M = 22.69, SD = 2.63). Psychology students received course credits for their participation, while the other students were paid an expense allowance of 20 €. The study was approved by the local ethic committee of the Faculty of Psychology at the Ruhr-University Bochum, and the Declaration of Helsinki was followed.

4.2.2. Experimental procedure

Participants were randomly assigned to either TSST or f-TSST. After signing informed consent, they filled in the SIAS (Mattick & Clarke, 1998) in the preparation room. Current affect was rated by filling in the PANAS (Watson et al., 1988) while the first saliva sample was delivered. Subsequently, participants were led into a testing room with either the TSST evaluation committee or the friendly f-TSST team, without previously having been informed about the condition. A fixed amount of an odour concentration, methyl benzoate (Sigma-Aldrich Corp., St. Louis, Missouri, USA), was dispersed during the participants' speech (Wiemers et al., 2014) using a compact ventilation device (TaoMaus, TAOASIS GmbH, Detmold, Germany). Back in the preparation room, participants delivered the first post saliva sample (+1 min) and then filled in the PANAS. Before the first startle block was initiated, a third saliva sample (+10 min) was collected. Participants then randomly started with either an olfactory or a visual startle block. Afterwards, the last saliva sample (+35 min) was delivered. For memory assessment, participants were instructed to freely recall as many objects present in the testing room as possible. Additionally, they engaged in an object recognition task,

rating on a 6-point scale how sure they were to have previously seen the objects displayed in the testing room.

Finally, participants rated six odours for their pleasantness on a 4-point scale (1 = "very pleasant", 2 = "pleasant", 3 = "unpleasant", 4 = "very unpleasant") and were asked to decide which of the six odours had been present during their speech or social interaction, respectively (*Figure 4.1.*).

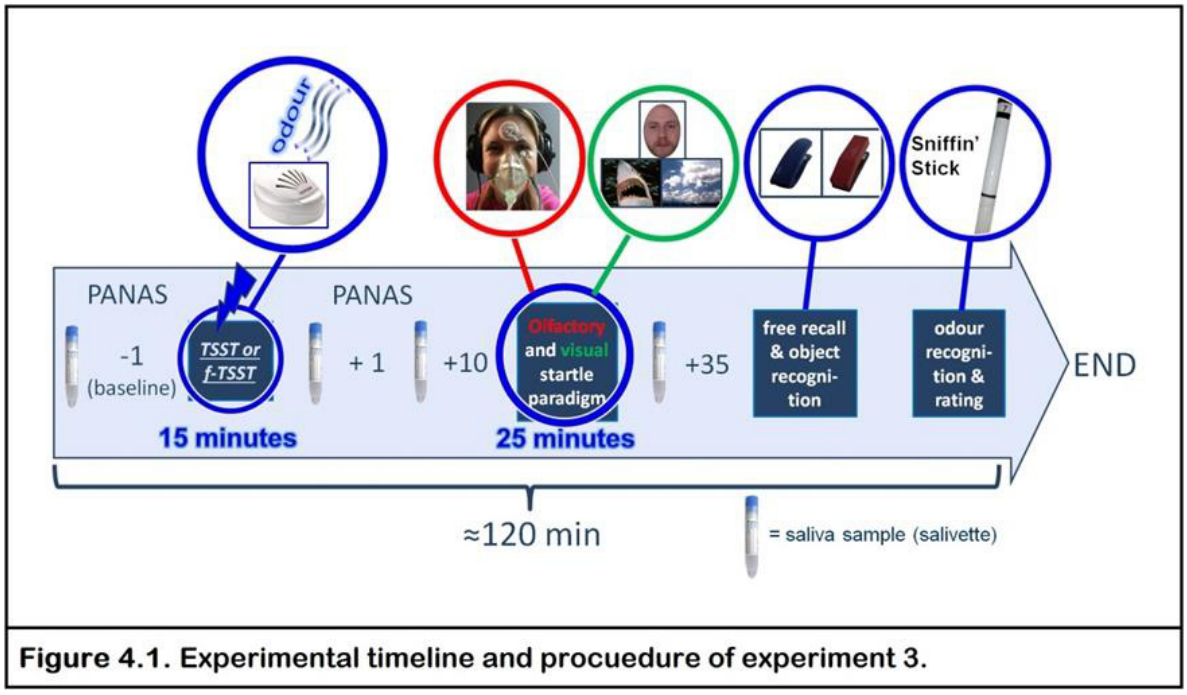

Figure 4.1. Experimental timeline and procuedure of experiment 3.

Besides the target odour methyl benzoate, two neutral unknown distractor odours presented during the startle session (bornyl acetate and linalool), as well as the unknown odour damascenone, and the known fragrances lemon and lavender (Sulmont et al., 2002) were part of the forced choice selection. Duration of the experiment was approximately 2 hours.

4.2.3. Material

4.2.3.1. Stress and control procedure

Trier Social Stress Test (TSST)

Stress was elicited using the modified version of the TSST (Kirschbaum et al., 1993; Wiemers et al., 2013). Please see section 1.1.2. and 2.2.3.1. for a detailed description of the procedure.

Friendly TSST (f-TSST)

As a control condition the f-TSST (Wiemers et al., 2012) was used as described in section 2.2.3.1.

4.2.3.2. Physiological stress measures

Salivary cortisol

In advance to the appointment, participants were instructed to refrain from any drug intake and physical exercise 24 hours before testing as well as drinking nothing but water and not brushing their teeth one hour before. Four saliva samples were delivered by each participant and deep-frozen at -18 °C immediately after testing. The cortisol analysis was performed at the local laboratory of the Ruhr-University Bochum with the *DEMEDITECs Cortisol Free in Saliva enzyme-linked immunosorbent assay (ELISA) Kit*. The resulting intra- and inter-assay CVs were below 10%. Due to the circadian rhythm of cortisol release, as described in section 1.1.1., participants were assigned to two time slots, either from 10 a.m. to 12 p.m. or 12 p.m. to 2 p.m. Assignment was done pseudo-randomly to assure equal distribution of the sexes and the two conditions over the two time slots.

Salivary alpha amylase

For measuring the response of the sympathetic nervous system, the enzyme α-amylase (sAA) was analysed from the saliva samples (Rohleder & Nater, 2009; Rohleder et al., 2004). The substrate CNP-G3 was used for measuring the enzymatic action of sAA at 405 nm. Intra- and inter-assay CV were both below 8%.

4.2.3.3. Affect measurement

Affect ratings were provided using the PANAS (Watson et al., 1988), as described in detail in section 2.2.3.3. Affect is calculated as a positive (PA) and a negative affect (NA) score.

4.2.4. Social Interaction Anxiety

To control for social interaction anxiety, the SIAS (Mattick & Clarke, 1998) was applied, as described in section 3.2.4. In this sample, an acceptable reliability derived with Cronbach's α = .710.

4.2.5. Startle evocation

The startle stimulus consisted of a 100 dB white noise with 50 ms duration and an instantaneous rise time, randomly presented via 80 Ω headphones (DT770M, beyerdynamic GmbH & Co. KG, Heilbronn, Germany). In contrast to experiment 2, the startle procedure included only one block with pictures and odours presented intermittently. This was done in order to avoid that the oxygen mask would be a confounding variable in modality comparison, as now participants wore the mask during both picture and odour presentation. At the

beginning of the startle block, 7 startle stimuli were presented during the 30 sec habituation phase. Between 0.5 and 2.5 sec after presentation onset of odour or picture the startle stimulus was applied. Every picture or odour was paired with the stimulus, but only in 50% of the presentations. Each visual or olfactory stimulus was presented twice for 3 seconds. During the ISI, which lasted for 8.5 sec, 0 to 2 startle stimuli were randomly applied.

4.2.6. Data recording

The electromyography recordings were conducted by means of two bio potential electrodes (EASYCAP GmbH, Herrsching, Germany) attached to the orbicularis oculi muscle of the left eye as described in section 1.5. (Blumenthal et al., 2005; Fridlund & Cacioppo, 1986; Lang et al., 1990). Additionally, a disposable ground electrode (GOLMED GmbH, Weddel, Germany) was attached to the forehead. For amplifying and transmitting the signal, the MP150 data acquisition device (BIOPAC Systems Inc., Essen, Germany) with filter settings 10 to 500 Hz was used. The startle programs were synchronised and executed with the software MatLab (version R2012a, MathWorks Inc., Ismaning, Germany).

4.2.7. Startle stimuli

4.2.7.1. Olfactory stimuli

Since one of the intentions of this experiment was to extend the findings of experiment 2 regarding acute stress effects, the same odours were used as in experiment 2. They had been chosen due to previous ratings as unfamiliar and neutral (Sulmont et al., 2002). The three essences methyl benzoate (60 µl), bornyl acetate (850 µl), and linalool (100 µl) (Sigma-Aldrich Co., Munich, Germany) were dissolved in 50 ml scentless paraffinum liquidum. These different odour concentrations were to assure comparable odour intensity (Wiemers et al., 2014). The odours were delivered by means of an in-house built 6-channel constant-flow (50 ml/s) olfactometer (Lorig et al., 1999) via oxygen masks (*ROESER Medical GmbH*, Essen, Germany) covering nose and mouth. In order to achieve maximum intensity of odour delivery at the moment of inhalation, the odour channel activation was adjusted to the olfactometer's mean latency (447.5 ms for onset, 608.5 ms for offset). For comparing startle responsivity on the basis of room air, five trials activating a channel containing a non-odorant cotton pad were included. Each odour was pseudo-randomly (never twice in a row) presented 7 times.

In accordance with experiment 2 (Herten et al., 2016), no breathing belt was implemented due to participants' compliance with breathing instructions (Adolph & Pause, 2012; Prehn et

al., 2006). The instructions consisted of a countdown (3 to 0) announcing participants to inhale when 0 is displayed, in synchronicity with the olfactometer's latency for odour onset.

4.2.7.2. Visual stimuli

As a variable for comparison, pictures were included in the startle block due to their known potency of modifying the startle response. This way, they provided reliable values to compare to startle responses to the odours. Pictures of unknown as well as the respective committee members known to the participant were included, as described in section 3.2.7.2. to assess modality effects associated with stress exposure. Each face was presented four times in order to assure valid trials with only two known committee members. Negative and positive stimuli of the IAPS (Lang et al., 2008) were the same as described in experiment 2, matched for arousal within each category and adapted from a previous startle study (Bradley et al., 2001; Herten et al., 2016) as described in section 3.2.7.2. To ensure comparison with olfactory stimuli, each picture was presented twice, but only once in combination with a startle stimulus. The presentation screen measured 15" x 12" and had a resolution of 1280 x 1024 pixels with a brightness of 100. During the ISI, a 20 x 20 pixels fixation cross was displayed at the centre of the screen. Participants were seated in an office chair at an approximate distance of 45 cm from the screen.

4.2.8. Startle data processing

Identification of valid startle trials was done with a semiautomatic mechanism of the software BrainVision Analyzer 2.0 (Brain Products GmbH, Gilching, Germany). Hereby, peak detection was set to a time range of 50 ms to 225 ms from startle onset. Before, a 50 Hz notch filter and a baseline correction mechanism (0-50 ms) were applied and the output was rectified. Verification and revision were done manually. In total, 3.2% of the startle responses were rejected, due to reactions outside the usual time scope (0.0613%) or non-responsiveness (amplitude did not exceed largest baseline amplitude by factor 2; 3.13%; Adolph & Pause, 2012).

4.2.9. Memory assessment

For assessing differences in memory performance between the groups, a free recall task and an object recognition task were applied as described in detail in section 2.2.6.

4.2.10. Statistical analyses

For startle analyses, mean values for each stimulus (three odours, four picture types) and sensory modality (olfactory, visual) were calculated. A repeated-measures ANOVA with within-subjects factors MODALITY (odours, pictures) and between-subjects factors CONDITION (stress, control) and SEX (male, female) was performed. It was repeated separately for the two modalities, with ODOURS (3) x CONDITION (2) x SEX (2) or PICTURES (4) x CONDITION (2) x SEX (2), respectively.

For memory data, mean values were calculated separately for stress and control group and for central and peripheral objects. In case of violation of the normal distribution, the data were log-transformed. To compare memory performance between stress and control group in the object recognition task, a discrimination index (DI) was calculated as described in detail in section 2.2.7. A repeated-measures ANOVA was performed with within-subjects factor object CATEGORY (central, peripheral) and between-subjects factors CONDITION (stress, control) and SEX (male, female) for both free recall and object recognition task. All statistical analyses were performed with SPSS 20.0.0.

4.3. Results

4.3.1. Participants

Of the 70 participants 2 were excluded due to software failures of the startle program, 3 participants were startle non-responders, and one expressed an enlarged startle amplitude of more than three SD from the mean. Another 5 exclusions concerned participants with unusually high baseline cortisol levels (> 3 SD from the mean), 4 exhibiting a cortisol decrease in the stress group, and one due to technical issues with the electrodes. Since cortisol as a measure of HPA activity is a questionable mediator between stress and the startle eye-blink response (section 3.4.1.), milder cortisol criteria than in experiment 2 were applied in favour of statistical power[3]. Of the 54 participants remaining, 26 were in the stress and 28 in the control group. As for menstrual cycle phases, 10 of the 26 female participants were in their luteal, 8 in their follicular phase, and 8 were ovulating. The Chi-Square test checking for differences in distribution of the different cycle phases between the groups showed no significant differences ($\chi^2(2) = .248$, $p = .884$).

[3] When applying the same 2.5 nmol/L cortisol responder criterion, the data show no changes in characteristics to the data reported here.

4.3.2. Physiological stress measures

Salivary cortisol

Since the cortisol data lacked normal distribution, they were log-transformed. A violation of sphericity was revealed by Mauchly's Test ($\chi^2(5) = 44.269$, $p < .001$), hence Greenhouse Geisser corrected p-values ($\varepsilon = .645$) are reported. An ANOVA with within-subjects factor TIME (baseline (-1), +1, +10, +35) and between-subjects factors CONDITION (stress, control) and SEX (male, female) was conducted. It showed that stress induction was successful, with participants of the stress group expressing an increase in cortisol concentration, reflected in a significant CONDITION x TIME interaction ($F(1.94,85.13) = 33.129$, $p < .001$). There were significant salivary cortisol differences between stress and control group at time points +1 ($t(51) = -2.666$, $p = .010$), +10 ($t(50) = -4.938$, $p < .001$), and +35 ($t(51) = -4.919$, $p < .001$), with maximum difference occurring at time point +10 (*Figure 4.2.*).

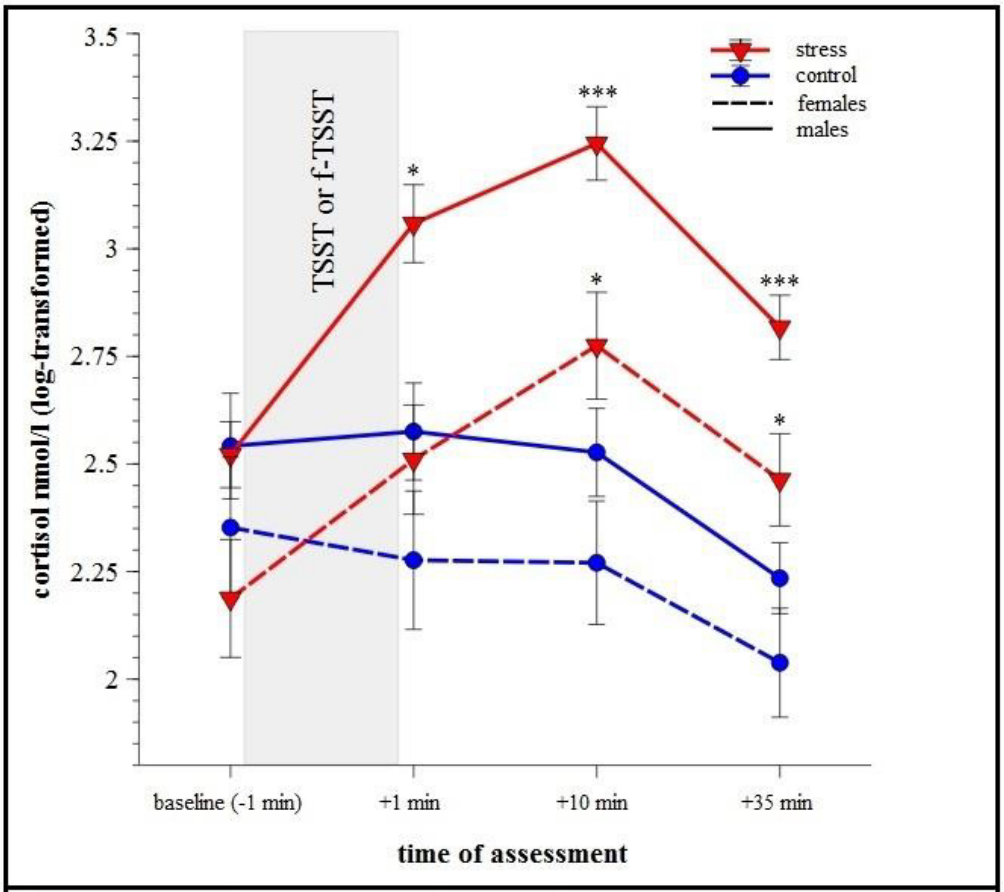

Figure 4.2. Time course of log-transformed cortisol data for both groups and sexes. Error bars refer to standard error of mean. Significances concern group differences.*$p < .05$,***$p < .001$.

A significant main within-subjects effect of TIME ($F(1.94,85.13) = 29.297$, $p < .001$) and significant main between-subjects effects of CONDITION ($F(1,44) = 7.979$, $p = .007$) were detected. Women exhibited slightly lower cortisol levels than men, which was expressed in a significant between-subjects effect of SEX ($F(1,44) = 8.512$, $p = .006$).

Salivary α-amylase

The sAA were log-transformed as they lacked normal distribution. Mauchly's Test showed a violation of sphericity ($\chi^2(5) = 6.178$, $p < .001$), thus Greenhouse Geisser corrected p-values ($\varepsilon = .789$) are reported. The ANOVA with within-subjects factor TIME (baseline (-1), +1, +10, +35) and between-subjects factors CONDITION (stress, control) and SEX (male, female) detected a significant within-subjects effect of TIME ($F(2.37,104.1) = 40.456$, $p < .001$), with a peak of sAA release one minute after the TSST/f-TSST, declining steadily in the aftermath in both groups (*Figure 4.3.*).

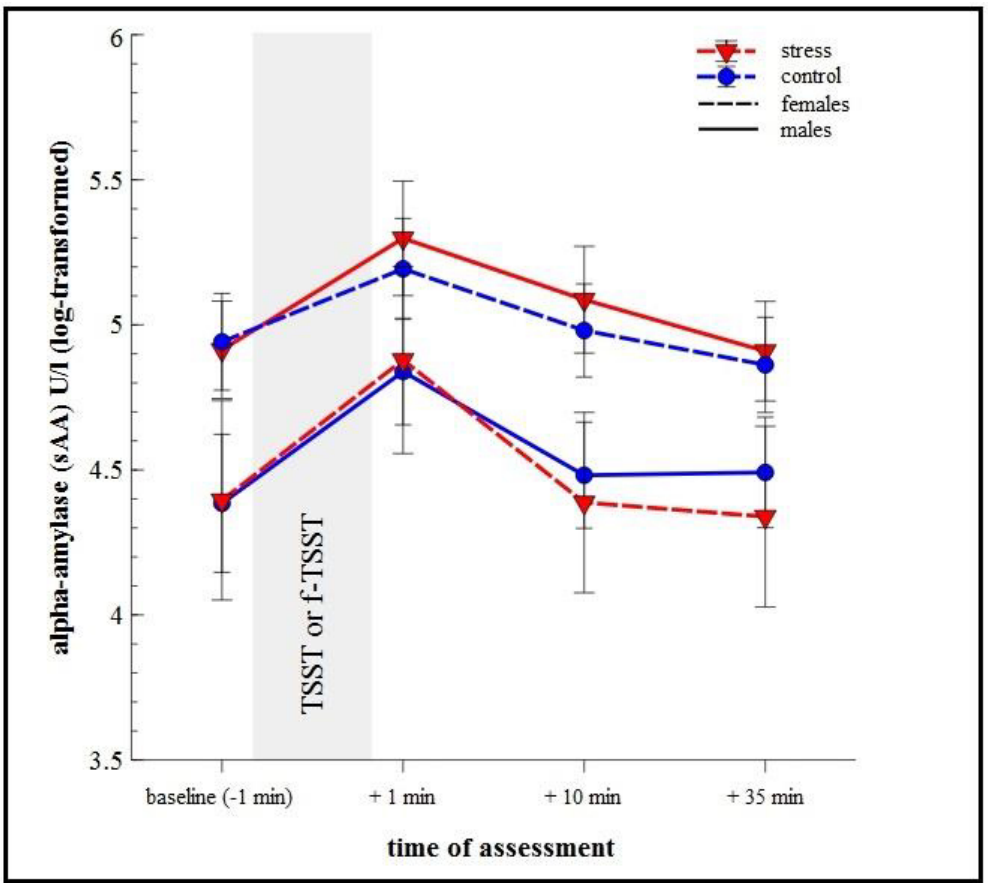

Figure 4.3. Time course of log-transformed sAA data for both groups and sexes. Error bars refer to standard error of mean.

No significant main effect of CONDITION ($F(1,44) = .024$, $p = .878$) and no CONDITION x TIME interaction ($F(2.37,104.1) = 1.188$, $p = .317$) were found, indicating a similar time course of sAA release in participants of both groups. In females, the sAA level of participants of the control group was slightly higher than in the stress group, whereas in males the opposite pattern was shown, reflected by a significant between-subjects CONDITION x SEX interaction ($F(1,44) = 4.126$, $p = .048$).

4.3.3. Affect measures

The PANAS scores show no group differences in affect ratings before the experimental manipulation. Participants of the stress group reported lower positive affect (PA) and higher negative affect (NA) compared to the control group after the experimental manipulation (*Table 4.1.*). A repeated-measures ANOVA with within-subjects factor TIME (pre, post) and between-subjects factors CONDITION (stress, control) and SEX (male, female) was conducted separately for the two affect scales (PA, NA).

Negative affect (NA)

The ANOVA for NA resulted in a significant within-subjects effect of TIME ($F(1,50) = 8.918$, $p = .004$) and a significant CONDITION x TIME interaction ($F(1,50) = 26.490$, $p < .001$), with the control group exhibiting lower post-assessment scores for the NA compared to pre-assessment scores, whereas the opposite pattern was shown in the stress group.

mean (SD)	stress	control
NA-pre	12.77 (3.80)	12.14 (1.78)
NA-post	18.23 (6.59)	10.68 (0.91)
PA-pre	29.50 (4.49)	28.54 (7.00)
PA-post	29.38 (5.93)	33.39 (7.54)

Table 4.1. Mean and standard deviation (SD) for negative affect (NA) and positive affect (PA) before (pre) and after (post) the experimental manipulation.

Moreover, a significant main effect of CONDITION ($F(1,50) = 25.743$, $p < .001$) was detected. Participants stressed had significantly higher scores at post-assessment of NA than

the control group ($t(52) = -6.005$, $p < .001$), whereas no differences were shown at pre-assessment ($t(52) = -.785$, $p = .436$).

Positive affect (PA)

Participants from the control group exhibited a higher PA after the experimental manipulation, reflected in a significant within-subjects effect of TIME ($F(1,50) = 12.712$, $p = .001$). Moreover, the analysis revealed a significant CONDITION x TIME interaction ($F(1,50) = 14.583$, $p < .001$). At post-assessment, participants of the control group showed a significantly higher PA than participants stressed ($t(52) = 2.160$, $p = .035$), whereas at pre-assessment there were no group differences ($t(52) = -.597$, $p = .553$).

4.3.4. Social Interaction Anxiety

No differences in SIAS scores between stress and control group were found ($F(1,52) = 2.704$, $p > .10$).

4.3.5. Startle responses

Descriptively, the startle responses of stressed participants in general are more pronounced than those of control participants (*Figure 4.4.)*. As the startle data were not normally distributed, they were log-transformed for further analyses. Mauchly's Test resulted in a violation of sphericity ($\chi^2(27) = 53.600$, $p = .002$), thus Greenhouse-Geisser corrected p-values ($\varepsilon = .785$) are reported. An ANOVA including the four factors MODALITY (odours, pictures), STIMULUS (3 odours, 4 pictures), CONDITION (stress, control), and SEX (male, female) revealed a significant main within-subjects effect of MODALITY ($F(1,50) = 8.305$, $p = .006$), with a higher startle responsivity for odours than for pictures ($M = .323$, $SD = .21$; $M = .300$, $SD = .21$).

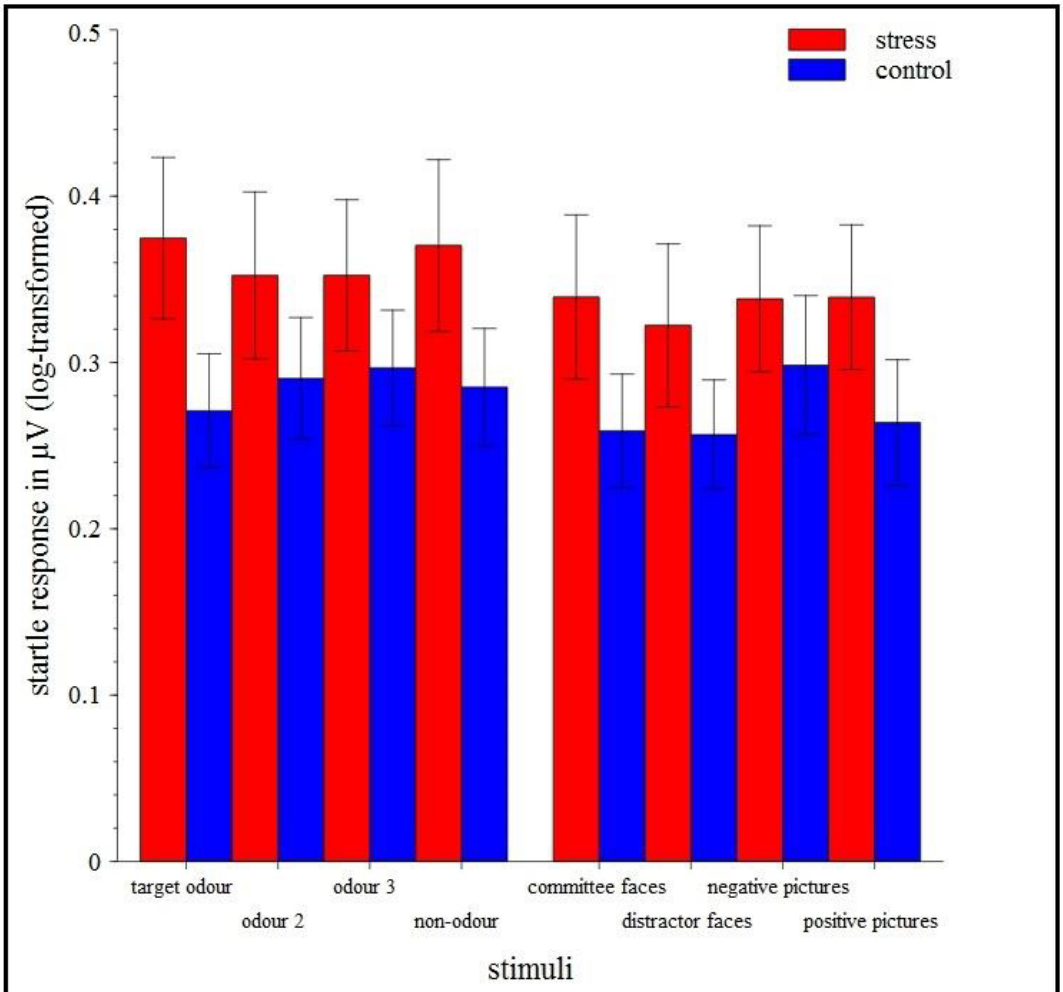

Figure 4.4. Mean log-transformed startle responses in both modalities (odours and pictures) for both groups. Error bars represent standard error of mean.
target odour = methyl benzoate, odour 2 = bornyl acetate, odour 3 = linalool, non-odour = cotton pad

Neither a significant main effect of CONDITION ($F(1,50) = 1.682$, $p = .201$), nor a CONDITION x STIMULUS interaction ($F(5.49,274.66) = 1.209$, $p = .303$) were found. There was a trend towards a threefold CONDITION x MODALITY x STIMULUS interaction ($F(3,150) = 2.413$, $p = .069$). A post-hoc independent-samples t-test for group comparison of startle in response to the different odours showed a trend towards higher startle responsivity to the target odour in stressed participants ($t(52) = -1.768$, $p = .083$). Startle amplitudes in response to the non-odour showed no significant differences to the other odours in the groups.

4.3.6. Odour recognition and odour ratings

Of participants from the stress group, 53.8% (14 out of 26) identified the target odour correctly out of 6 odours included in a forced choice trial. In contrast, only 25% (7 out of 28)

of the control participants recognised the target odour. This group difference showed a trend towards significance ($\chi^2(1) = 2.645$, $p = .104$).

The subjective ratings of the three odours were compared in a repeated-measures ANOVA with the factors CONDITION (stress, control) x ODOUR (1 target, 2 distractor odours) x SEX (male, female) to test for group differences in judgement of the affective quality of the target odour. No significant CONDITION x ODOUR interaction ($F(3,150) = .372$, $p = .773$), and no significant main effect of CONDITION ($F(1,50) = .255$, $p = .616$) were detected.

4.3.7. Memory performance

Free recall

A repeated-measures ANOVA was conducted with within-subjects factor object CATEGORY (central, peripheral) and between-subjects factors CONDITION (stress, control) and SEX (male, female).

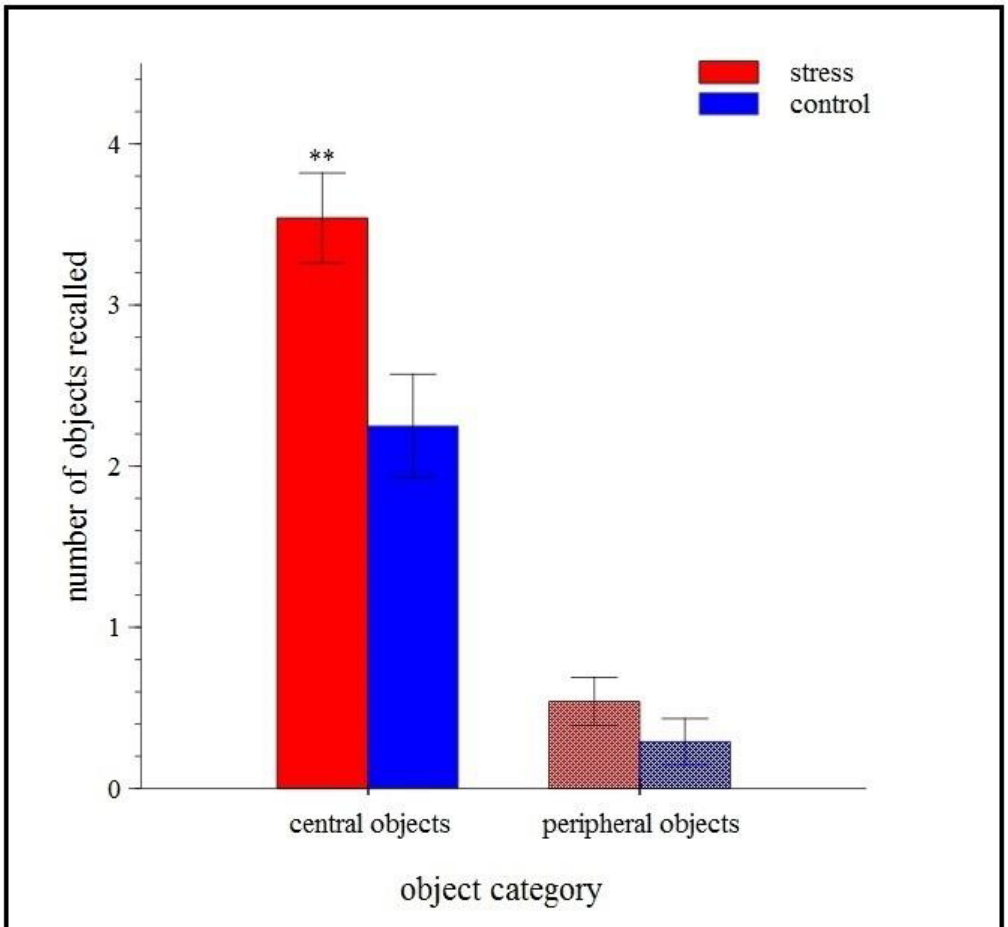

Figure 4.5. Mean number of central and peripheral objects named in the free recall task in both groups. Error bars refer to standard error of mean. Significances concern group differences.** $p < .01$.

A significant CONDITION x CATEGORY interaction was shown for the free recall task ($F(1,50) = 4.937$, $p = .031$, $\eta^2 = .090$), as well as a main effect of CONDITION ($F(1,50) = 9.265$, $p = .004$, $\eta^2 = .156$), with participants stressed demonstrating a generally better memory performance than control participants, particularly for central objects (*Figure 4.5.*). A significant within-subjects effect of CATEGORY was shown ($F(1,50) = 121.082$, $p < .001$, $\eta^2 = .708$), as both groups showed a better memory performance for central than for peripheral objects.

Discrimination Index (DI)

The repeated-measures ANOVA for the DI as calculated from the object recognition task resulted in a significant main effect of CONDITION, revealing a better memory performance in the stress than in the control group ($F(1,48) = 5.193$, $p = .027$, $\eta^2 = .098$).

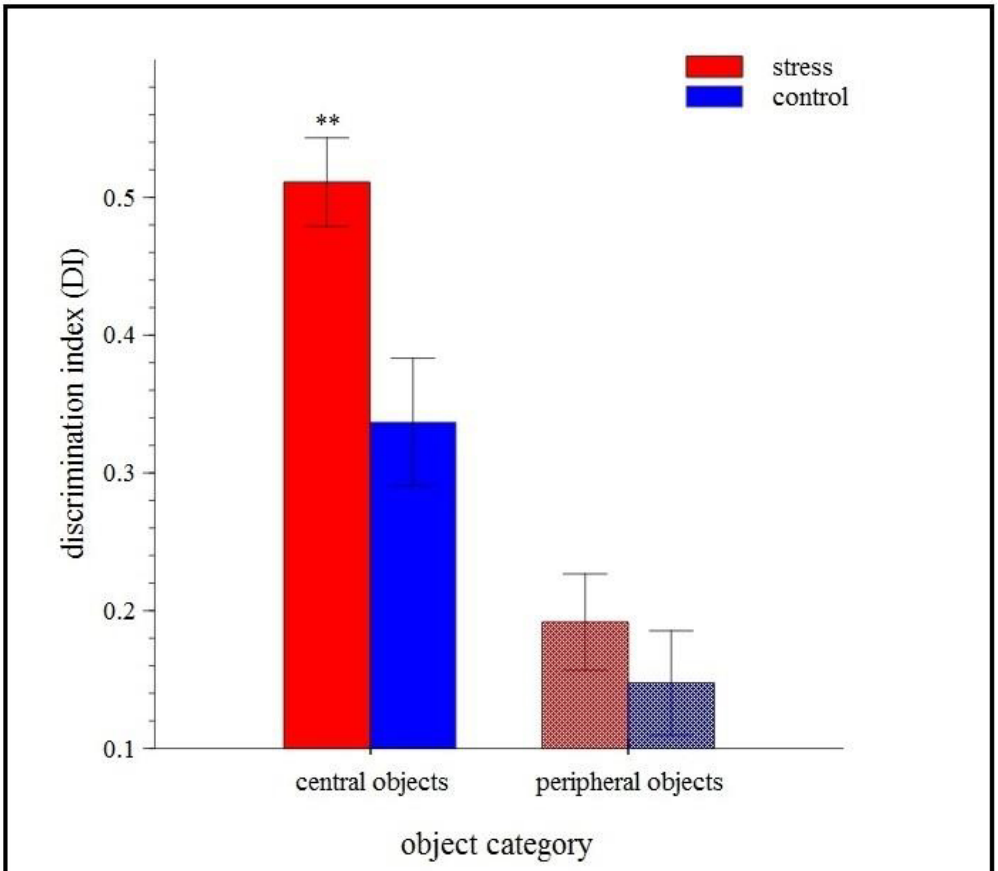

Figure 4.6. Discrimination Index (DI) for central and peripheral objects recognised by stress and control groups in the object recognition task. Error bars refer to standard error of mean. Significances concern group differences.**$p < .01$.

This effect was particularly pronounced for central objects, reflected in a significant CONDITION x CATEGORY interaction ($F(1,48) = 5.532$, $p = .023$, $\eta^2 = .103$) (*Figure 4.6.*). A significant main effect of CATEGORY ($F(1,48) = 84.507$, $p < .001$, $\eta^2 = .638$) shows the stress effect to be more pronounced for central than for peripheral objects.

4.4. Discussion

The current study on human startle responsivity and memory for a stressful situation in its immediate aftermath showed that stressed participants tended towards higher startle responsivity to the target odour. Stressed participants additionally exhibited descriptively higher general startle responsivity across all stimuli and modalities compared to control participants. Moreover, they showed a trend towards better recognition of the target odour and significantly enhanced memory performance for the central objects of the stressful situation.

4.4.1. Stress induction

4.4.1.1. Physiological stress measures

The cortisol time course of the groups, with stressed participants exhibiting a cortisol rise peaking at +10 minutes, whereas control participants show a decline, indicates a successful stress induction. The physiological response in terms of cortisol and sAA is of the same characteristics as previously shown with this paradigm (Wiemers et al., 2012). The sex differences are assumed to be due to a previously observed lower cortisol responsivity in women in confrontation with the TSST (Kirschbaum et al., 1999), as described in section 1.1.1., and do not lead to sex differences in the dependent variables.

4.4.1.2. Affect measures

The affect measures, in particular the higher post-assessment NA score in stressed and lower score in control participants, confirm subjective stress induction.

4.4.2. Startle responsivity

Descriptively, stressed compared to control participants show enhanced startle responsivity in immediate aftermath of the stress experience, in contrast to the findings of Deuter et al. (2012) describing an even diminished startle response. Their findings are based on the CPT as a stressor which lacks the social component of the TSST. Thus, the results lack comparability. Apparently, the increased vigilance caused by stress (section 1.1.) leads to a more pronounced startle response in order to protect the organism from potential threat. Since the CPT elicits a

stress response solely based on physiological stress due to triggering the organism's temperature regulation system, the response differs from a situation of psychosocial stress including ego threat.

The hypothesis that stressed participants show enhanced specificity, expressed in a stronger startle response to the target odour, was confirmed at a trend level. As startle responsivity in general was increased, this contrasts the findings of experiment 2 of enhanced responsivity at the expense of specificity (Herten et al., 2016). Since the startle paradigm occurred in closer temporal proximity to odour exposure, implicit and explicit memory for it was stronger, which might have mediated enhanced responsivity to the target odour. Since stress was found to potentiate sensory input (Munk et al., 1996), experience of the odour in the acute stress phase was probably intensified.

In line with experiment 2 and a previous study comparing these modalities (Adolph & Pause, 2012), startle responsivity for the olfactory modality was higher than for the visual modality. As explained in section 4.2.5., participants were wearing the oxygen mask during presentation of stimuli in both modalities in the current experiment. Thus, it can be ruled out that it was responsible for enhanced responsivity to odours compared to pictures in experiment 2. The olfactory system's strong bond with the cortical nucleus of the amygdala explained in sections 1.3. and 1.4. might cause this effect (Doty, 2001). It is conceivable that rapid responses in areas of the amygdala elicited by odours led to a state of increased vigilance for an amygdaloidal response cascade initiated by the startle stimulus.

4.4.3. Subjective odour ratings and odour recognition

As hypothesised, recognition of the target odour was superior in the stress contrasting the control group, at a trend level. This supports the notion of potentiated odour experience in the acute stress phase, as discussed in the previous section. In the immediate aftermath of the stressful episode including the odour, explicit memory for its aspects are enhanced. This demonstrates the modified attentional and short-term memory aspects under stress before consolidation processes come into play.

In contrast to this, the odour ratings between the groups did not differ – the target odour was not rated more negative than the distractor odours. This indicates that implicit memory, accounting for a more aversive affect in connection with the odour experienced during the stressful situation, does not immediately benefit from the stress effects. Since the odour ratings differ after one day, as shown in experiment 2, implicit memory might be boosted with consolidation processes, causing the odour ratings to become increasingly negative with time.

4.4.4. Memory measures

The memory effects found 24 hours after stress induction in experiment 2 as well as previous studies (Wiemers et al., 2013, 2014) apparently are present already in the immediate aftermath of the stressful experience. Consolidation thus does not seem to be the main process accounting for these effects. As described in section 1.1., glucocorticoids acting on the noradrenergic system of the basolateral amygdala facilitate the beneficial effects of stress on memory consolidation (Roozendaal, Okuda, de Quervain, & McGaugh, 2006). However, rapidly proceeding non-genomic effects on immediate attentional and mnemonic processes in the acute stress phase (de Kloet et al., 2005) apparently are pivotal for increased memory as found in experiments 1 and 3. In line with this, increased vigilance under stress has been shown to create a vantage point for processing of stressor-related information of relevance (Hermans, Henckens, Joëls, & Fernández, 2014; Ramos & Arnsten, 2007). In a stressful situation, attention and memory for stimuli of potential relevance for similar future situations is exceptionally promoted (de Kloet et al., 2005). Objects used might be linked to the specific situation to potentially reoccur in the future and are thus encoded and as a result remembered better, even before being consolidated into long-term memory storage.

4.4.5. Conclusion

The results of this experiment show that at a trend level psychosocial stress enhances the auditory fear-potentiated startle response in the acute phase, apparently due to increased vigilance triggering the organism's protection mechanisms. This effect was shown particularly in response to the odour ambient during the stressful episode, indicating increased response specificity. The enhancing stress effects on memory are detectable in the immediate aftermath of the psychosocial stressor, even before salient enough stimuli are then being transferred into long-term memory.

5. General discussion

The investigation reported in this dissertation was conducted in three experiments to shed light on whether stress influences

- fixation behaviour, hereby mediating memory outcome,
- odour perception and memory combined with startle responsivity after 24 hours,
- odour perception and memory in combination with startle responsivity and object memory in immediate aftermath of psychosocial stress.

In this section, the findings of the three experiments are integrated for drawing a final conclusion on the topics the dissertation aimed to contribute to. The general discussion is overarching with regard to similar findings of the three experiments as well as concerning the different parameters assessed.

5.1. Summary of the main findings

5.1.1. Experiment 1

The findings of this experiment show a significant enhancement of stress on fixation duration and number of fixations on central objects. At the same time, fixation on the faces of the committee members was more pronounced in the control than in the stress group, presumably due to gaze aversion of stressed participants towards the aversive source of stress. Even though the beneficial effects of stress on recognition memory and free recall after 24 hours could be replicated, fixation and memory measures did not correlate, nor did fixation mediate the effects of stress on memory. The results indicate that modified fixation patterns under stress cause advantageous stimulus processing potentially influencing the selection of stimuli of relevance to subsequently be consolidated into long-term memory. Nevertheless, they show that there is no direct and no strong relation between fixation measures and memory outcome under stress.

5.1.2. Experiment 2

In this experiment, stress effects on startle responsivity, affective olfactory perception and odour memory 24 hours after stress exposure were investigated. The results show generally enhanced startle responsivity 24 hours after psychosocial stress, combined with a decrease in specificity. The odour ambient during the stressful episode tended to be rated more aversive, however, recognition memory of and startle responsivity to the odour were not increased. Apparently, the stress experience leads to increased vigilance and response sensitivity to aversive stimuli as assessed by the startle paradigm on the next day. This effect might be

based on a shift in amygdala processing towards increased anxiety, causing a "spill-over" effect with regard to responsivity at the expense of specificity.

5.1.3. Experiment 3

Similar to experiment 2, stress effects on startle response specificity to an odour ambient during the stressful episode were assessed in immediate aftermath of the psychosocial stressor. To expand the findings of experiment 1, object memory was assessed in the acute stress phase. The results show enhanced startle response specificity under stress with an increased response to the odour present during the stressful experience, at a trend level. Stressed participants further tended to recognise the odour better than control participants. Memory for central objects was found to be significantly increased in the stress group. The findings suggest that stress exerts immediate influence on startle responsivity and in particular memory processes, before consolidation processes come into play.

5.2. Stress induction

All three studies used a modified version of the TSST for stress induction (sections 1.1.2. and 2.2.3.1.), in comparison to the f-TSST as the control condition (section 2.2.3.1.). By eliciting psychosocial evaluative threat, establishing a motivated performance, and integrating the aspect of uncontrollability, the TSST leads to a strong stress response in the laboratory. However, it has to be noted that, in comparison to natural situations like accidents, the stress response elicited by the TSST in terms of cortisol release and affective reaction is of a rather moderate nature and by no means comparable to a traumatic event. The use of a responder criterion thus provides a tool to distinguish stressed from non-stressed participants within the given range of cortisol rise elicited by the TSST. If the investigation focusses on HPA axis activation, a stringent responder criterion, as used in experiment 2, ensures that all participants in the stress and no participant in the control condition exhibit a stress response in terms of cortisol release triggered by HPA axis activation. As the influence of stress on fixation behaviour was not believed to be mainly based on HPA axis activation – actually, a stronger association with the rapidly increasing sAA response would have been more conceivable – the responder criterion in experiment 1 differed from that used in experiment 2; only responses distinctively deviating from the mean of the respective group were excluded. This criterion was adopted in experiment 3 for increased statistical power. The exclusions in all three experiments were comparable to the number of excluded participants in previous studies with the TSST (e. g. Kirschbaum et al., 1993).

In all three experiments, the TSST successfully elicited stress by means of HPA axis activation, expressed in stronger cortisol elevation, as well as increased negative affect after the experimental manipulation. Hereby, the main source of social-evaluative threat and thus of stress was the committee. Due to their reserved and cold behaviour, the context of the job interview becomes its meaning and is perceived as stressful, despite the knowledge about it being a simulated situation. Sex differences in cortisol release as observed in experiment 1 were due to intake of hormonal contraceptives (Kirschbaum et al., 1999; Nielsen et al., 2013) and did not cause other sex differences in the dependent variables assessed. Neither did the sex differences in cortisol release found in experiment 3, which are presumed to be due to dampened cortisol release in women, as previously observed in laboratory stress situations such as the TSST (Kirschbaum et al., 1999). At the same time, the f-TSST provided a well-controlled and comparable control condition not activating the HPA axis, whereas SNS activity in terms of a rise in sAA is comparable in stress and control group.

5.3. Fixation and attention

Experiment 1 aimed at establishing a link between fixation measures and memory performance in stressed participants. That is, to investigate whether memory enhancement in stressed participants is mediated by longer and more fixations on the objects memorised. As hypothesised, stressed participants exhibited significantly prolonged fixation times and more frequent fixations on the central objects than control participants in experiment 1. Thus, stress exerts influence on fixation behaviour. Despite the missing correlation and mediation between fixation measures and memory outcome on the next day in connection with stress, fixation patterns seem to have an impact on information encoding and, in concert with attentional processes, contribute to optimal selection and processing of relevant input. In line with this, stressed participants in experiments 1 and 3 demonstrate significantly enhanced memory for central objects. The presence of these effects in the immediate aftermath of the stress experience in experiment 3 shows that processes of attentional narrowing towards potentially relevant items have occurred. Since stressed participants did show different fixation patterns from control participants, these are obviously associated with the attentional modification which apparently accounted for the narrowing of focus on the central objects. As discussed in section 2.4.5., the impact of this immediate effect might decrease with time and with increasing action of consolidation processes, which would be the reason why no direct correlations could be detected. Moreover, it was only assessed whether participants did or did not remember the objects, but not memory for details. Longer and more frequent fixations

would however suggest a better encoding of and thus memory for details, whereas less attention towards an object does not necessarily mean that it is not going to be remembered, but memory for details might be weak. Since lowest level representational structures of objects are formed within a few hundred milliseconds of fixation (Rensink & Enns, 1998), features, for instance the object category, can be processed rapidly. However, focussed attention is needed to form a coherent and more detailed object representation, as low level structures are fragile and may be overwritten by new concurring input (Rensink, 2000a, 2000b). Memory for details is thus more likely to be correlated to fixation times. In experiment 1 the memory effects were assessed with an "all-or-nothing" method, especially with regard to the free recall which showed the strongest effects. Assessment of the number of details remembered in an additional memory task might demonstrate a closer relationship between fixation and memory.

Since the fixation measures *fixation count* and *average fixation duration* showed enhanced expression from the moment of object usage, it can be confirmed that the categorisation of the objects based on their use by the committee members is reasonable. As the committee members are the main threatening stimulus in the TSST, their behaviour, including the items they involve, are bound to the main stressor and thus become of relevance themselves. Hence, the categorisation as central refers to the role of the item in the situation and its potential importance. This shows that location is not the main factor regarding the meaning of an object as was previously referred to for categorisation as central (Wessel, De Kooy, & Merckelbach, 2000). Previous studies have categorised central and peripheral items on the same basis as in the current dissertation (Peth, Vossel, & Gamer, 2012; Wiemers et al., 2013). Their results suggested this categorisation to be well-founded. However, the current investigation is the first to introduce eye tracking measures to validate this categorisation and expand the related findings to attention and fixation measures.

On the basis of these findings it could be shown that the effect of stress on fixation measures, as the case for the memory parameters, was particularly significant for central objects. This might be due to more stimulus related bottom-up processing under stress. As previously described in section 2.1., stress leads to dominance of bottom-up processes, e. g. in attentional selection (Buschman & Miller, 2007), whereas top-down processes are curtailed (Arnsten, 2009; Sänger et al., 2014). Since top-down control relies on prefrontal areas (Buschman & Miller, 2007), this effect might be due to strong inhibition of prefrontal activation under stress as found in an fMRI study (van Stegeren, Roozendaal, Kindt, Wolf, & Joëls, 2010). Volitional direction of focus and thus attention as a result of top-down selection

for both locations and objects are thought to rely on neural circuits in prefrontal cortex (Desimone & Duncan, 1995; Kastner & Ungerleider, 2000). With stress impairing functioning of the prefrontal cortex (Schwabe & Wolf, 2010), attention is thus drawn on basis of bottom-up processes towards more specific, situationally relevant aspects. As the central objects are being used by the main stressor, the committee, they gain relevance in this specific situation and thus are prioritised. This mechanism is mainly dependent on the amygdala which enhances perception and representational activation of stimuli of emotional relevance (Anderson & Phelps, 2001). In visual scene perception stimuli compete with each other for focussed attention. It was shown that arousal increases mutual inhibition effects between these competing stimuli (*see for review*: Mather & Sutherland, 2011). Neutral stimuli competing for attention can either be impaired or enhanced in dependence of their priority, which is defined by stimulus based perceptual salience (bottom-up) or attentional focus through motivational, goal-directed top-down processes, respectively. Particularly negative emotional arousal increases attentional selectivity (Sutherland & Mather, 2012). Hence, in a situation of high arousal, the selection of stimuli of relevance becomes more precise, as representations of equal priority might all be suppressed. A more sensitive detection of differences in relevance between the stimuli is thus promoted by amygdaloidal processes. Perceptual thresholds in the respective sensory brain areas are lowered, facilitating enhanced sensory processing (Davis & Whalen, 2001). In line with this, the amygdala was found to be activated as a moment-to-moment representation of affective experience related to the stimulus perceived, subsequently leading to memory enhancement dependent on emotional intensity of the respective stimulus (Canli et al., 2000). Amygdaloidal processes thus seem to contribute to enhancement of the signal-to-noise ratio for relevant and central aspects versus peripheral aspects. The process of competition for representation just described is initiated with perception and ongoing up to subsequent long-term consolidation processes (Mather & Sutherland, 2011). These findings indicate that arousal, as elicited by stress, but also caused by affective features of the stimulus itself, modifies fixation and attention processes which in successive mechanisms cause selective items to be consolidated and remembered. Experiment 1 demonstrates modified fixation patterns under stress which seem to be beneficial for later stages of stimulus processing. Even though a direct correlation between fixation and memory is lacking, subsequent mechanisms causing the memory enhancement under stress can only process what earlier sensory stages provided them with.

In contrast to the object fixation data, the fixation measures on the faces of the committee members show the reversed pattern, with stressed participants fixating the faces less often and

for shorter times than control participants. In situations of social evaluative threat, the eyes of the persons involved were shown to be the strongest fear-inducing feature (Öhman, 1986). This makes gaze avoidance a common and successful strategy for reduction of discomfort in a social stress situation (Posner & Rothbart, 2000). In addition to this affective component, findings demonstrate an association of psychophysiological stress measures with gaze avoidance, as a quadratic relation with cortisol concentration was found in children (de Veld, Riksen-Walraven, & de Weerth, 2014). Especially in socially anxious individuals direct gaze is perceived as a threat, causing its avoidance (Horley, Williams, Gonsalvez, & Gordon, 2003; Wieser, Pauli, Alpers, & Mühlberger, 2009). As fear of social evaluation has been identified as the main component of social anxiousness (Kocovski & Endler, 2000), participants under socially evaluative threat, as elicited by the TSST, are very likely to avoid direct eye contact with the committee members. Fixation measures of experiment 1 can confirm these findings and expand them in terms of behavioural data of a controlled laboratory investigation.

5.4. Memory

Experiment 1 sought to replicate the findings of enhanced memory performance 24 hours after a stressful experience (Wiemers et al., 2013) to investigate whether this effect is mediated by fixation behaviour on the objects encoded and memorised. This investigation was expanded to memory effects in the acute stress phase in experiment 3. Both experiments assessed object recognition memory via a computerised task as well as free recall for the objects involved. Memory results of experiments 1 and 3 confirmed previous findings of memory enhancement under stress, and experiment 3 added to the question whether these effects occur already after a short delay or are mainly based on consolidation processes, promoting better memory performance on the next day.

Stress-induced memory enhancement

In all three experiments, stress was induced with the TSST. Memory encoding thus took place during stress exposure. This way, close temporal proximity of the affective and hormonal stress response with stimulus encoding did occur, which is essential for successful memory enhancement under stress, as described in section 1.2. (Henckens et al., 2009; Joëls et al., 2006; Lupien et al., 2002; Roozendaal, 2002; Roozendaal & McGaugh, 2011). Besides, it was previously shown that encoding is exceptionally successful when the to be remembered input is in close association with the stressor (de Kloet et al., 1999). Since in the TSST the encoded objects are directly related to the stressful source, the committee members handling them, this facilitating effect was thus given in all experiments of this dissertation. Moreover, stress

intensity and memory enhancement are generally assumed to maintain an inverted U-shaped relationship (section 3.1.). That is, very low levels of stress and thus cortisol concentration as well as very strong stress responses do not promote memory enhancement, while moderate stress does so. The TSST usually elicits medium levels of stress – in contrast to real-life experiences such as an accident – with moderate cortisol increases, which is optimal for memory effects. The optimal requirements for the memory promoting effects of noradrenergic activity in basolateral amygdala enabling the effects of GR activation in hippocampus, as described in section 1.2. (Roozendaal, 2003), were thus fulfilled.

Time course of memory enhancement

Whereas experiment 1 shows effects on delayed recognition and free recall memory performances 24 hours after the stressor, results of experiment 3 imply that these effects are already present in the immediate aftermath of the stressful experience, circa 40 minutes afterwards. During this relatively short time span, encoding and selection processes of the stimulus representations take place, but memory for them is not yet stabilised by consolidation processes. Nevertheless, after a short delay, memory for selective items is already enhanced by stress. Thus, it can be concluded that consolidation processes are not essential for detection of stress effects on memory enhancement. The main contribution of consolidation processes might lie in further enhancing the signal-to-noise ratio in a way that more relevant information becomes an even stronger memory trace, whereas irrelevant information, which has not proved to be central, gets overwritten. Supporting this notion, data of experiment 1 shows much stronger effects of group differences in free recall for the Condition main effect (η^2 = .489) as well as the Category-Condition interaction (η^2 = .365) 24 hours after the stress experience compared to the immediate aftermath as assessed in experiment 3 (η^2 = .156 and η^2 = .090), despite the smaller sample size in experiment 1. Thus, the differences in memory performance between the groups are more pronounced 24 hours later, but already present in the direct aftermath. Additionally, the effect is even more pronounced for central objects on the next day, as shown by the Category-Condition interaction. In relation to the fixation findings, this implies that modification of processes during encoding is a main contributor to stress effects on memory enhancement of objects in the TSST, as already discussed in section 5.3.

Differential memory processes

In section 5.5., olfactory memory is discussed to rely on rather implicit memory processes in contrast to encoding and consolidation of the visual stimuli used in the experiments of this

thesis. To further compare olfactory to visual memory processes as assessed with the two different memory tasks used in this thesis, the differences between the memory processes underlying free recall and object recognition which have been found in experiments 1 and 3 are discussed and related to implicit and explicit memory processes in the following section.

Since free recall is based on recollection rather than familiarity, the stronger stress effects in this task compared to the object recognition task in experiment 1 confirm previous results of particularly enhanced recollection under stress (Wiemers et al., 2013). However, there seems to be a dissociation effect regarding the different forms of memory in immediate aftermath (experiment 3) versus 24 hours after stress (experiment 1) – data of the object recognition task show a stronger effect for the Condition-Category interaction than results of the free recall task in experiment 3. This indicates that free recall memory (recollection-based) and memory in the object recognition task (familiarity-based) are differently influenced by acute stress effects and by consolidation effects after stress during the course of one day. In line with these findings, results from an event-related functional MRI study suggest that emotion effects on retrieval memory are based on recollection rather than familiarity, in particularly after lengthy (1 year) retention intervals (Dolcos, LaBar, & Cabeza, 2005). Moreover, this study could show more pronounced activity in amygdala, entorhinal cortex, and hippocampus when emotional visual stimuli were correctly remembered than the case for neutral visual stimuli. In addition, only in amygdala and hippocampus, not in entorhinal cortex, this influence of emotional stimuli was superior for recollection compared to familiarity. Thus, the effect of emotional stimuli seems to particularly influence recollection memory via enhanced activation of amygdala and hippocampus. Not only for emotional stimuli, but also for emotional contexts and neutral stimuli within these contexts, a dominance of recollection over familiarity has been found. During retrieval of neutral objects that had previously been associated with an emotionally laden (negative or positive) context, a late central-parietal old/new difference in event-related potential (ERP) of 400 to 700 milliseconds was shown (Ventura-Bort et al., 2016). This describes a positive maximum in parietal ERP modulation which has its onset at around 400 to 700 milliseconds after stimulus presentation when correctly identifying the respective item. Centro-parietal waveforms for old/new differences in this range (> 500 ms) have previously been found to reflect recollection-based remembering (Rugg & Curran, 2007). The association with this ERP in the investigation of Ventura-Bort and colleagues, in combination with the effects of stress on particularly recollection-based memory performance as previously found in other studies and the current dissertation, indicate an influence of stress particularly on recollection of affective

stimuli. Recollection memory is thought to be based on more effortful processes, rather slowly accessing conscious information about the respective object and its context (Rugg & Curran, 2007). Moreover, recollection memory benefits from meaningful encoding and requires more attention, both during encoding and retrieval, compared to familiarity (*see for review*: Rugg & Yonelinas, 2003). This fits well with the findings of enhanced fixation measures under stress as shown in experiment 1, reflecting more elaborate attentional processes in stressed compared to control participants. It further underlines the role of central objects as their utilisation is goal-directed and thus promotes encoding within a meaningful context in contrast to the peripheral items. Recollection memory is further assumed to be a continuous process inducing a mnemonic state after a certain threshold for memory retrieval strength has been exceeded (*see for review*: Yonelinas, 2002). As discussed in this and the previous section, stress promotes sensitivity in selection of stimuli of potential relevance to be successfully perceived, encoded, and finally consolidated. Threshold dependency of recollection memory would thus explain why this form of recognition memory is more affected by stress and arousal, leading to more pronounced memory differences between stress and control group.

Central versus peripheral object memory

In line with previous evidence (Kensinger, 2009; Mather, 2007; Waring & Kensinger, 2011; Wiemers et al., 2013), experiments 1 and 3 both confirm the effect of enhanced memory in stressed participants particularly for central objects. As described in section 5.3., the categorisation of the items as central and peripheral objects is underlined by the fixation data; central objects are fixated longer and more often than peripheral objects, in particular by stressed participants. This has been shown to be the case after the central objects had been used by the committee members, further validating this categorisation. Hence, even though it were office items not emotionally laden in nature, they gained an emotional valence through their association with the main stressor, the committee. In line with this, it has formerly been shown that initially neutral elements are remembered better through pairing with an emotionally charged context (Ventura-Bort et al., 2016). In this thesis the effect was shown with a relatively moderate stress induction contrasting experiences like accidents or attacks. For persons experiencing a traumatic event, the effect of previously neutral objects which are encountered during the experience to become associated with the trauma is even stronger. Thus, neutral objects can cue strong emotional memories through this previous association and can be subject to fear generalisation. Given that the emotional context, which in the TSST are the committee members, confers affective qualities on the items experienced within this

context, these objects are subject to emotional binding processes promoting their encoding as well as memory consolidation. That is, arousing stimuli lead to affective responses promoting their binding to the context (*see for review*: Mather, 2007), which in the current investigation is the situation of socially evaluative threat. In addition, cortisol has been found to enhance item-context binding of central memory contents, suggesting to serve a protective function against memory generalisation (van Ast, Cornelisse, Meeter, & Kindt, 2014). The items are thus associated with and bound to the stressful situation by being used by the committee members, as also supported by the findings of the fixation data discussed in the previous section.

Stress did especially promote memory for central items, but also enhanced memory for peripheral items, in the free recall task in experiment 1 even significantly (section 2.3.3.). These findings contrast those of a previous study reporting a memory trade-off effect under emotionally arousing conditions, with better memory for central aspects while memory for peripheral aspects was impaired (Mather, 2007). A possible explanation for these contradictory findings might refer to the cognitive demands of the free speech task in the TSST, leading to fewer resources available for memory encoding. The suppression of the emotion-induced trade-off effect by inclusion of a cognitively demanding secondary task has been addressed before (*see for review*: Kensinger, 2009). A further possibility could be of methodical nature, as studies reporting the trade-off effect made use of emotionally arousing stimuli capturing attention, such as pictures of car accidents. It was demonstrated that in studies without "attention magnets" the emotion-induced trade-off effect could not be replicated in favour of generally improved memory rather than memory narrowing (*see for review*: Laney, Campbell, Heuer, & Reisberg, 2004). Since none of the items used in the TSST can be regarded as an "attention magnet" for they are not emotionally arousing or salient in nature, but only gain their relevance through becoming related to the main stressor, this could explain why the data does not replicate any memory trade-off effects of stress.

5.5. Olfaction

Experiments 2 and 3 both investigated whether a stressful experience influences the perception of a previously unknown and neutral odour towards a more negative rating of and enhanced memory for the odour, and thus a fear-potentiated startle in response to this odour, 24 hours after or in the acute stress phase, respectively.

For experiments 2 and 3 mixed results were found with regard to recognition of the odour as well as its rating. The target odour was recognised by 53.8% of the stressed participants in

experiment 3 – in the immediate aftermath of the stressful experience – compared to only 10% of correct identifications after 24 hours, in experiment 2 (Herten et al., 2016). In experiment 3, in closer temporal proximity to the experimental manipulation, stress and control groups differed at a trend level (53.8% versus 25% of correct identifications), unlike on the next day where the exact small amount of participants (two) in each group recognised the target odour. The data further show a dissociation of subjective rating of and memory for the target odour – whereas acute stress effects seem to be beneficial for memory of the odour present, it does not influence its subjective rating; one day later, though, explicit memory for this odour apparently declines in parallel to consolidation of the affective response eliciting a more negative rating (Herten et al., 2016). Different forgetting rates of explicit and implicit memory contents might be responsible for this dissociation. Explicit and implicit memory processes have been shown to act independently of each other (Cowan & Stadler, 1996; Graf & Schacter, 1985; Roediger, 1990; Schacter, 1987). Explicit memory might decrease more rapidly than implicit memory, as was previously found particularly for fear-related memory contents (Packard, Rodríguez-Fornells, Stein, Nicolás, & Fuentemilla, 2014). This could have caused the more negative rating of an odour whose conscious association with the stressful experience on the previous day has ceased. In the light of these findings, the memory processes discussed in the previous section suggest a relation of the different time courses of recollection and familiarity with implicit and explicit memory processes. Models on these different forms of memory have suggested that familiarity and implicit memory both underlie the same main memory process (e. g. Mandler, 1980). It can be presumed that explicit memory is rather associated with recollection, whereas implicit memory affords cues which are related to familiarity judgements (*see for review*: Rugg & Yonelinas, 2003). Thus, the more pronounced impact of stress on recollection of the objects 24 hours later, as found in experiment 2, would suggest an improvement of explicit rather than implicit memory. This contrasts the notion that explicit memory declines more rapidly than implicit memory (Packard et al., 2014). However, this might be dependent on the modality of the stimuli encoded and retrieved. It is conceivable that visual stimuli form stronger explicit memory contents which are enhanced under stress and consolidated, such that their recollection is better after one day. Due to the lack of thalamic gating described in section 1.3., olfactory stimuli are prone to create strong implicit memory contents. This might explain the differential effects with regard to odour rating and odour memory, as implicit memory for the odour might have been consolidated, leading to a more negative rating on the next day. At any rate, odours have strong potency to become related to an experience, also due to their

connections to crucial emotion and memory structures like amygdala and hippocampus. A set of novel visual stimuli was shown to become associated with specific odours in a functional magnetic resonance imaging study, causing increased activation in right anterior hippocampus in contrast to 'visual-only' acquisition of those stimuli (Ghio et al., 2016). Enhanced activation of memory structures by olfactory stimuli might reflect a potential of odours to initiate additional memory processes, maybe of implicit nature. These and the findings of experiment 3 indicate a high amenability of the olfactory system to become related to new memory contents.

It was hypothesised in experiments 2 and 3 that the odour ambient in the testing room would later elicit an enhanced startle response due to a negative association with the odour present during the stressful experience. One day after stress exposure, this prediction could not be confirmed, and in the immediate aftermath only at a trend level. The explanation might be a shift towards enhanced responsivity at the expense of specificity, based on activity shifts in the amygdala, as previously discussed (section 3.4.2.). In this case, the increased vigilance would lead to a "spill-over" effect masking specific responses to the stimuli presented during the startle block. This alone would not explain why the same effect was shown not only for the acute stress phase in immediate aftermath of the stressor in experiment 3, but also in experiment 2, when the acute stress response had ceased. A possible explanation for this would be that context effects of memory of the stressful episode lead to re-experiencing the same internal state as on the previous day. Memory retrieval is known to be better when taking place under the same contextual circumstances as memory encoding, which does not only refer to the external environment (Godden & Baddeley, 1975; Schwabe & Wolf, 2009), but also to internal emotional states (*see for review*: Bookbinder & Brainerd, 2016). It was shown that odours have the potential of being efficient contextual cues for stress-related memory contents, as described in section 3.1. (Wiemers et al., 2014). Since stress has been found to enhance item-context binding (van Ast et al., 2014), context effects of memory encoding and retrieval are likely to be enhanced by stress. It would thus be conceivable that re-exposure to the odour during the startle session triggered the same affective internal state as on the previous day and thus lead to the same "spill-over" effect found before. The negative affect in participants of the stress group was not found to be increased after testing on the second day. However, the assessment was not done immediately after the startle block as it was on day one, instantly after stress exposure, but at the end of the testing session, approximately 35 minutes after the startle session. It thus remains unclear whether negative affect was enhanced in the stress group at the moment of startle exposure.

5.6. Startle responsivity

Experiment 2 and 3 were designed to show that stress leads to enhancement of startle responsivity, particularly with regard to an odour experienced during stress induction, 24 hours later or in immediate aftermath of the stressor, respectively. Since the startle reflex is a protection mechanism against potential threat to the organism (section 1.5.), increased vigilance induced by stress is bound to enhance this reflex. For the acute stress phase this was found in experiment 3, even though only at a trend level. Interestingly this effect was still shown one day later in experiment 2, after cessation of the acute psychophysiological stress response. The raw startle response voltage was found to be slightly higher in the acute stress phase, as shown in experiment 3, than 24 hours later according to the results of experiment 2 (Herten et al., 2016). The more prominent group differences in this experiment are due to a more pronounced startle response in the control group in direct aftermath of the f-TSST, which might be explained by SNS activation due to the social interaction. As already discussed in section 2.4.1.1., the f-TSST as a control condition also leads to sAA increase (Wiemers et al., 2013, 2012), indicating similar SNS activation as in stressed participants. Noradrenergic activation reflecting SNS activity has been shown to lead to sensitisation of the acoustic startle response in rodents (Fendt, Koch, & Schnitzler, 1994). In humans, the startle response was also found to be highly sensitive to arousal (Balada et al., 2014). Thus, arousal in control participants in experiment 3 could have caused enhanced startle responsivity immediately after the control procedure, whereas in experiment 2, SNS activity on the second day had ceased. This might explain the more pronounced differences between stress and control group found in experiment 2 compared to the acute phase in experiment 3. Experiment 2 is the first investigation to show enhanced human startle responsivity one day after stress exposure and demonstrates a long-term effect of the TSST on increased vigilance, even in the absence of an acute psycho-physiological stress response.

As part of the hypothesis of experiment 2 and 3, startle response specificity was predicted to be enhanced under stress, such that the startle amplitude in response to the target odour methyl benzoate should be higher than in response to the two distractor odours bornyl acetate and linalool. Experiment 2 did not confirm this expectation, but in experiment 3 this was found at a trend level. It was assumed that this result had not been found 24 hours after stress induction in experiment 2, since enhanced responsivity was accompanied by reduced specificity due to a shift of amygdala functioning. However, besides increased specificity, startle responsivity in general was increased in immediate aftermath of the stressor in experiment 3, which contrasts the assumption made for the results of experiment 2 claiming

enhanced responsivity at the expense of specificity (Herten et al., 2016). As previously described in sections 1.1. and 5.3., stress leads to increased bottom-up habitual at the expense of top-down controlled stimulus processing (Buschman & Miller, 2007). This mechanism might have contributed to more stimulus-based responsivity during the acute stress phase in experiment 3 and thus to more pronounced startle amplitudes in response to the odour just experienced during the stressful episode. In contrast to this, the cessation of the acute stress response might have caused elimination of the odour aversion. This assumption would be supported by findings of a study on taste-potentiated odour conditioning, showing that extinction of the previously conditioned taste aversion thoroughly extirpated the potentiated odour aversion of the paired olfactory stimulus (Hatfield & Gallagher, 1995). In addition to this, participants of the stress group were being debriefed about nature and purpose of the psychosocial stressor, with the committee behaviour being standardised and by no means related to their performance, after the first day's testing session in experiment 2. This knowledge about the previous stressor might have influenced the startle outcome on the second day, similar to the experiment of Hatfield and Gallagher demonstrating this extinction in the olfactory modality. Since the startle response is a very sensitive measurement tool, it is possible that the target odour as an aversive stimulus was extinguished by the knowledge about the stressor in combination with the diminished psychophysiological stress response of the acute stress phase on the previous day. Contrasting this, stressed participants in experiment 3 were not debriefed until after completion of the experimental session. Thus, they were still of the belief that the voice and video recordings taken of them would be used for further analysis by a committee they have just had an unpleasant encounter with, in addition to the acute psychophysiological stress response causing the increasingly promoted stimulus-response processing. In addition to this, odours are thought to be more dependent on the original encoding context than stimuli in other modalities, mainly due to their non-lexical nature (Hinton & Henley, 1993).

It was shown in experiments 2 and 3 that odours seem to have a special potency to trigger strong responses under stress-induced increased vigilance, expressed in a significantly more pronounced startle response than the case for visual stimuli. In experiment 2 participants were wearing oxygen masks solely for odour delivery and not during the visual startle block. To exclude this being a confounding variable causing the increased startle response for the olfactory modality, a single startle block with visual and olfactory stimuli presented intermittently was applied in experiment 3. Participants thus were wearing the oxygen mask during both visual and olfactory stimuli presentation. Experiment 3 confirmed the finding of

more pronounced startle responsivity to olfactory than to visual stimuli which had apparently not been caused by the oxygen mask. This finding is in line with a previous investigation's showing higher startle magnitudes in response to odours than to pictures (Adolph & Pause, 2012). Furthermore, in direct comparison of responses in three modalities, olfactory, visual and lexical, odours have been found to elicit qualitatively and quantitatively different responses with the largest amount of affect (Hinton & Henley, 1993). Once again these effects can be related to amygdala activation. Results of a Positron Emission Tomography (PET) study demonstrate that pleasant and unpleasant affective feedback in response to stimuli in any sensory modality relies on activation of the same core network consisting of the orbitofrontal cortex, the temporal pole, and the superior frontal gyrus, each of the left hemisphere (Royet et al., 2000). However, only judgements on pleasantness of stimuli in the olfactory modality led to additional increase in regional cerebral blood flow in the amygdala. This finding of superiority of the olfactory system in activating the amygdala by odours of affective valence in direct comparison to visual and auditory stimuli is supported by the results of experiments 2 and 3 of this dissertation, demonstrating more pronounced startle responsivity to olfactory than to visual stimuli. It has to be noted, though, that the startle reflex is designed to provide the organism with a fastest-response overall-protection mechanism, soon as danger is detected in any sensory modality. By this signal-integration of the different sensory modalities, a clear separation of the potencies of these modalities can hardly be made, since the evolving startle response might be a potentiation of multiple inputs to the different channels. However, the lack of specificity thought to be related to amygdaloidal shifting processes is more pronounced in response to the odours – in both experiments 2 and 3 – than to the pictures, as the negative pictures and the pictures of known in contrast to unknown committee members still elicit higher startle amplitudes than the positive pictures in stressed participants, reflecting the typical fear-potentiated startle response in experiments 2 and 3. This would also support the assumption of special potency of olfactory stimuli in enhancing the startle amplitude and thus leading to stronger responsivity than visual stimuli. In the acute stress phase, the emphasis of the effect on olfactory stimuli seems to be even more pronounced, as stressed participants exhibit a slightly lower specificity in response to pictures, contrasting a slightly higher specificity in response to odours, while the opposite pattern was found 24 hours later in experiment 2. The main findings discussed here, compared to similar findings of previous studies are summarised in table 5.1.

effect	supporting findings	consistent results of this thesis
narrowing of focus towards significant items and selective attention under stress and arousal	improved attentional selectivity in stressed participants (Chajut & Algom, 2003); more attention to central details in the emotionally arousing condition (Christianson et al., 1991)	enhanced fixation on central items in stressed participants in experiment 1
memory enhancement under stress for central information	better memory performance in stressed participants for central objects (used by committee) in the TSST (Kensinger, 2009; Mather, 2007; Waring & Kensinger, 2011; Wiemers et al., 2013)	experiment 1 and 3 demonstrate a better memory in the stress group in particular for central items, in both a free recall and an object recognition task
no correlation of fixation times and memory outcome	stimulus exposure time does not predict memory performance when fixation time is partialled out (Loftus, 1972)	despite enhanced fixation and memory measures in stressed participants in experiment 1, they did not correlate
covariation of memory performance with fixation duration	better memory for central details in emotional condition in subjects with equal number of eye fixations on that critical central detail (Christianson et al., 1991)	both memory performance and fixation duration varied dependent on group membership in experiment 1
enhanced startle eye-blink response 24 hours after stress exposure	a one-time stress experience had an influence on eye-blink conditioning one day later in male rats (Beylin & Shors, 1998)	the 15 minutes TSST elicited a stress response strong enough to enhance general startle responsivity one day later
more pronounced startle eye-blink reflex in response to odours than to pictures	odour presentations during emotion regulation were accompanied by larger startle magnitudes than picture presentations (Adolph & Pause, 2012)	both the stress and control group exhibit more pronounced startle responsivity to odours than to pictures in experiments 2 and 3

Table 5.1. Main findings of the experiments in this thesis and related findings from previous studies.

5.7. Clinical implications and practical application

In many countries, particularly in the USA, eye witness reports are crucial for evidence and their testimonies can have serious consequences, especially when death penalty is an option. Due to their relevance, they exert a literally pivotal influence on the decision of the judge and jury and thus the impact of their misidentifications contributes to a great extent to wrongful convictions (National Academy of Sciences, 2014). Experiment 1 has shown that stress, which is experienced in even more extreme forms in witnesses of crime or accidents, influences fixation behaviour. Although this is a functional mechanism in order to focus on potentially significant features in this specific situation for an appropriate response, this does not warrant flawless encoding and, above all, retrieval of the information in an eye witness situation. Besides suggestive questioning and other confounding factors, study 1 of this thesis has shown that stressful situations influence fixation measures associated with different attentional processes. Additionally, it was demonstrated that there is no direct translation of fixation behaviour into memory outcome. It has to be considered that eye witness reports might be produced on basis of effects such as weapon focus and item-context binding. As previously discussed (section 5.4.), an "attention magnet" can lead to an emotion-induced trade-off effect at the expense of memory for details not related to the central item (Mather, 2007). It was shown that scenes involving weapons lead to attentional narrowing, as previously described in this thesis (section 2.1.), so that eye witnesses focussing on the weapon can divide less attention to other details of the scene leading to a poor memory for crucial details of the crime, for instance the culprit's face or appearance (E. F. Loftus, Loftus, & Messo, 1987; Maass & Köhnken, 1989). Results of experiment 1 indicate that a stressful context has the potency of lending initially non-arousing items an affective character. Thus, in a situation of high arousal and stress, context effects like this might lead to trade-off effects even for stimuli non-salient in nature. Context effects, as discussed in this thesis (Godden & Baddeley, 1975; Schwabe & Wolf, 2009), may furthermore lead to more accurate identifications when potential suspects are being presented to the witness in the context of the crime. Knowledge gained from results of psychological investigations like this has been applied and institutions do already consider phenomena like weapon focus and context effects, for instance by showing witnesses pictures of suspects at the crime scene (National Academy of Sciences, 2014). Eye tracking research with participants under stressful conditions and with simulated crime scenes would certainly add to the understanding of processes eye witnesses go through during their encoding experience and when retrieving the information.

Other forms of emotional memory are thought to rely on emotion-induced context binding, such as "flashbulb memories". Most people have found themselves in a situation where they heard about a strong emotionally arousing event, such as the 9/11 incident. Experiences like this lead to strong memories for location, people's company and other details of the occasion people first heard of the incident. However, it was shown that these putatively strong memories are mainly based on a higher confidence of accurately remembering the event in every detail, rather than proving to be factually different to memories of other non-emotional events (*see for review*: Hirst & Phelps, 2016). This underlines the finding in experiment 1 about a lack of one-to-one correspondence between emotional memory encoding and retrieval processes, which is also relevant for eye witness cases. Flashbulb memories are for instance strongly influenced by consolidation and rehearsal processes. With every recall of the event, the memory is bound to be modified by other witness and media reports, not necessarily in the accurate direction. Adaptive mechanisms like the stress response do thus not always turn out to be functional.

A further issue demonstrating this is the pathology of PTSD, a condition caused by extremely stressful, often highly traumatic experiences. It is characterised by re-experiencing the event in terms of intrusive memories and intense dreams. Often, these re-experiences are triggered by cues which can occur in any sensory modality like odours, flashes, pictures, and loud noises and can take forms of intense flashbacks leading to hallucinations and illusions. As part of the pathology, patients are in a state of increased arousal with hypervigilance leading to an exaggerated startle response (e. g. Butler et al., 1990). These symptoms demonstrate a binding of the experience to the original encoding context of the traumatic event, a mechanism which is believed to actually serve a protective function in order to avoid memory generalisation (van Ast et al., 2014). Part of this effect is believed to be based on heightened HPA axis in concert with SNS activity during the traumatic experience, influencing amygdaloidal and hippocampal processes in such intensity that a "failure to recover from a nearly universal set of emotions and reactions" (Yehuda, 2002) derives. This reflects the myriad processes under stress and concurrence of its various effects in the different sensory modalities, which unfortunately can have negative and dysfunctional consequences. A moderate stressor as used in the experiments of this thesis has shown that neutral items can become central through being associated with a stressful context (experiments 1 – 3) and that a negative perception of an odour involved in this situation can be induced (implicitly in experiment 2 and explicitly in experiment 3). Patients suffering from PTSD often associate stimuli perceived as neutral by other people with the negative

experience and thus perceive the stimuli itself as negative, such as an odour (Vermetten, Schmahl, Southwick, & Bremner, 2007). An approach to remediate or weaken the main symptoms of PTSD is to untie the fearful memories from their original and generalised context by re-exposure under protective circumstances or under administration of cortisol promoting new encoding. A thorough understanding of memory strengthening processes under stress, including all sensory modalities, can contribute to elaboration of the appropriate method to weaken or even cure PTSD symptoms.

5.8. Limitations

The TSST was shown to successfully induce stress in all three experiments, causing the expected cortisol response as well as increased negative affect in the stress group. Moreover, the f-TSST proved to be a suitable control condition, not activating the HPA axis and not leading to an increase in negative affect in the control group. Even though these two conditions are highly comparable, they are different in the nature of their respective social interaction – while the TSST is characterised by a social evaluative situation causing the participant to hold a monologue, the f-TSST is a friendly and casual interaction. While these features do not influence the main parameters assessed in experiment 2 and 3, they do have an influence on fixation measures in experiment 1. Since it is natural to keep eye contact and to more or less constantly try to read the facial expression of an interaction partner in a relaxed dialogue, participants of the f-TSST are expected to show more and longer fixations on the committee faces than stressed participants who tend to avoid gaze towards and eye contact with the committee as explained in sections 2.4.4. and 5.3. The motivation of participants of the control group to direct focus onto the committee members evidently distracts from directing their attention towards the objects. Although this difference is certainly owed to the stress response in the one group versus the more naturalistic non-stressful interaction in the other, it is rather the nature of the tasks causing the control participants to face towards the committee members, whereas the stress group avoids this and might even focus on the objects for reasons of emotion regulation. Nevertheless, also in non-stressful situations it is natural to scan the environment, and many natural, non-laboratory stress situations also involve social interactions, such that the data conducted in experiment 1 are still valid and representative. This is also shown in fixation measures for peripheral objects which demonstrate no significant differences between the groups. With regard to experiments 2 and 3, the differences in the nature of the two conditions do not seem to have influenced the startle

response to the committee faces, since no group differences in startle amplitude in response to the pictures of the committee known as well as unknown were found between the groups.

Since memory for the committee faces has not been assessed, experiment 1 cannot answer the question how the dissociation in fixation times on objects and faces between the groups did actually influence differences in memory performance. If the more pronounced fixation of the faces in the control group would have resulted in a better memory performance in comparison to the stress group fixating the faces less often, the association between the fixation measures and memory outcome would probably have been strengthened and might even have led to a correlation between the measures. This however remains unclear.

Since olfactory stimuli are likely to have access to implicit memory contents (sections 1.3. and 3.1.), and differences in object memory between free recall and object recognition have been observed in both experiments 1 and 3, it is discussed whether stress differentially influences free recall and object recognition memory, and – at the background of implicit olfactory memory as well as the rather implicit assessment of the startle paradigm – implicit and explicit memory processes. With regard to this, a differentiation between recognition and familiarity is discussed. Since free recall is un-cued memory retrieval, it is based on recognition rather than familiarity (section 5.4.) which is why this task has been included in addition to the object recognition task. Results of the object recognition task have however been dichotomised and not been analysed with a Receiver Operating Characteristic of the AUC, such that no clear distinction between recognition and familiarity can be made in the memory data of experiments 1 and 3.

Moreover, memory assessment with free recall is an "all-or-nothing" method, and object recognition memory has been analysed as "remembered" versus "not remembered". Thus, it is unclear whether stressed participants did also have a more detailed memory for the objects than control participants. Inclusion of this variable would have also provided a measure to be associated with fixation times, as it is conceivable that longer and more frequent fixation lead to better encoding of object features rather than just memory for the object in general.

The results in startle responsivity to the target odour show no (experiment 2) or only weak enhancement (experiment 3), respectively, in the stress group. In both experiments 2 and 3, no disgust-related or negative odours were included which would have provided a direct comparison to the valence of the three odours presented, reflected in the respective effects on the startle response. Thus, the data can neither provide a direct comparison of the effects of negative olfactory stimuli with the odours presented, nor a comparison of the effects of negative stimuli on the startle response between the two modalities. Thus, no conclusion can

be drawn on why the target odour did not lead to a stronger startle response in the stress group in experiment 2 and only a non-significant trend in experiment 3. Due to the significantly more negative rating of the target odour in the stress group as reported in experiment 2 as well as the trend towards an increase in startle responsivity to this odour in experiment 3, a lack of negativity associated with the target odour seems unlikely.

In experiment 2, baseline cortisol differences between stress and control group have been found. The ANCOVA has shown that these baseline differences, neither on the first nor on the second day, have not been responsible for the differences in startle responsivity as found on the second day. They might however be responsible for other differences between the groups, mediating the outcome in an uncertain way. The fact that the effect of enhanced responsivity in the stress group is even more pronounced when conducting the ANCOVA suggests a slight influence of the cortisol baseline difference. As far as could be assessed with trait questionnaires, the groups have been shown to be very similar. Of special interest in this regard are social interaction anxiety and neuroticism. Hence, the baseline cortisol differences could be due to for instance more participants of the stress group showing up too late for testing rather than personality differences. As mentioned before (section 3.4.1.1.), it is unclear whether cortisol influences startle responsivity at all. As it is not the only neuroendocrine factor composing the stress response, other factors the stress response consists of might be more crucial for influencing startle responsivity.

5.9. Future directions

As all our senses are finely adjusted to rapidly provide us with pivotal information needed to deal with challenges of everyday life as well as with extraordinary situations of extreme stress and threat, future research is needed to focus on the influence of stress on our sensory systems and adjacent brain regions and processes to add to our understanding of organisms under stress. This is especially important considering clinical conditions deriving from the growing demands of work life, increasing the rate of stress-induced diseases in the population.

Fixation and memory

Experiment 1 indicates, despite lacking correlations, an influence of stress on fixation behaviour and encoding processes. To further investigate potential interrelations between stress, fixation, and memory, an experimental manipulation of fixation patterns in a controlled setting would be necessary to draw conclusions about how the time spent fixating an object is associated with memory. This way, fixation times can be kept constant in both groups and the

direct influence of stress in terms of cortisol and affect can be assessed and related to the fixation times.

As previous studies have already shown (Wiemers et al., 2013, 2012; Wiemers & Wolf, 2015), the experiments in this thesis confirm that the friendly version of the TSST leads to a similar sAA secretion as the stress condition. Since the effects of sAA are more immediate in comparison to the effects of cortisol, a stronger association of sAA secretion with fixation measures can be assumed. Thus, a control condition not activating the SNS would shed light on the relation between SNS activity and fixation behaviour.

The effects of stress on memory enhancement were assessed 24 hours later in experiment 1, combined with findings of stress effects on enhanced fixation measures at the moment of stress. This difference in the assessment time of fixation and memory might be the reason for the lack of correlation between these variables. A direct association of fixation behaviour with short-term memory processes might be more likely than with regard to long-term memory processes. It would thus be interesting to assess fixation in combination with memory performance in immediate aftermath of the stress induction, as done in experiment 3, before consolidation processes come into play. Hereby, memory for details should be additionally assessed. It would also add to the comprehension of fear acquisition and trauma-related diseases such as PTSD to thoroughly understand encoding mechanisms under stress and their later representations in long-term memory. For this kind of pathologies, it would be of special interest to find out more about item-context bindings, such that these could be extinguished to avoid generalisation of fear and triggering of fearful memories by contextual cues. Evidence suggests that arousing stimuli trigger emotional reactions prioritising binding of those stimuli to their context (Hadley & MacKay, 2006; Mackay et al., 2004; MacKay & Ahmetzanov, 2005). Moreover, strong memory effects have been found for emotional items, particularly in combination with a stressful situation (Abercrombie, Speck, & Monticelli, 2006; Bradley, Greenwald, Petry, & Lang, 1992; Kuhlmann & Wolf, 2006). In the experimental setup used in this dissertation, inclusion of emotional and arousing items would provide a comparison to the neutral office items in terms of fixation times, memory strength, and temporal development of their representations from short-term to long-term memory storage. Even though central objects had the potential of becoming important in the stressful episode and lead to enhanced memory, they might not have exceeded a certain threshold (like e. g. a gun feasibly would have done) to demonstrate a direct correlation of fixation on and memory for them, since memory was basically assessed with “all-or-nothing” tasks. This could have been overcome by assessing memory for details of the objects. It is conceivable, that details of a gun are

remembered better than details of a ruler, as a weapon is more salient and emotionally arousing. Future investigations with this experimental setup should thus consider assessing memory for details of central and peripheral objects to have a more precise measure for comparing the memory performance of stressed and control participants and achieve a closer association with fixation times. Additionally, items of different categories could be included in future studies to investigate whether this would enhance more categorical memory contents in contrast to more item-specific memory contents. Further, the inclusion of emotional and neutral out-of-context items (e. g. a skull versus toaster) would be interesting for a direct comparison of the influence of an emotionally arousing, stressful context versus a non-stressful context on neutral versus arousing items, as well as the influence of arousing versus non-arousing stimuli on item-context binding.

For future studies in the field of stress, especially when investigating memory effects, fixation behaviour should always be taken into consideration. Hereby, also processes like top-down and bottom-up control, influencing scanning behaviour and the perception of items as salient or mundane, need to be considered. Also studies comparing the impact of stress on memory in adults and children (Quas, Rush, Yim, & Nikolayev, 2014) should include fixation behaviour as an additional measure to account for the different developmental stages of experience-related top-down and bottom-up influences in adults and children.

Finally, with expanding knowledge about fixation behaviour under stress, it could potentially provide a new measure of stress, in addition to heart rate and other biomarkers. Relating behavioural parameters like fixation measures to inner states is a useful and convenient measure that has been previously considered as such. As described in sections 2.4.4. and 5.3., gaze avoidance is considered as an expression of shame and embarrassment in several cultures (Edelmann & Neto, 1989). In terms of stress, fixation times would be a useful behavioural measure which is not intrusive, relatively easy to establish, and presumably very congruent and stable across different cultures.

Olfaction and startle responsivity

Due to their high potency to enter long-term memory storage in combination with very emotional memory contents (Herz, 1998; Herz & Cupchik, 1992, 1995; Herz & Schooler, 2002; Hinton & Henley, 1993), which is mainly due to their strong and direct connection to areas of the amygdala (Herz et al., 2004), odours should be increasingly integrated into future research. As the olfactory system is prone to trigger implicit processes, new study designs for detection of odour-related affective states and memories might have to be considered. Since the startle paradigm is very useful in doing so, a refinement of the startle setup and procedure

for odours should be implemented in future studies. Also concerning the stimuli, it would be helpful to further investigate the applicability of olfactory stimuli in laboratory set-ups, for instance with regard to being neutral and unknown. The odours delivered besides the target odour during the olfactory startle block in experiments 2 and 3 were included for comparison of startle responsivity to see whether the target odour elicits a stronger response. Therefore, the distractor odours were selected due to being neutral and unknown (Sulmont et al., 2002). However, to be able to judge whether the similarity of the startle responses to the three odours in experiment 2 (and only a trend towards stronger responsivity to the target odour in experiment 3) is based on lack in negative affect in response to the target odour or on a "spill-over" effect causing anxiety generalisation, a negative and a positive odour would have to be additionally included in future investigations.

In experiment 2, a strict cortisol responder criterion was applied to include only participants stressed based on HPA axis activation (Herten et al., 2016). Through this selection criterion, it could be assumed that the enhanced startle responsivity detected in the stress group was at least partly based on the increase in cortisol. However, to really investigate the influence of cortisol on startle responsivity, a pharmacological study manipulating the cortisol concentration by administration of hydrocortisone would reveal causal relations between the two measures. This would be helpful to investigate, to really gain knowledge about the influence of cortisol on startle responsivity which cannot be answered by experiment 2 and 3.

Besides the instant effects of acute stress on startle responsivity, investigating long-term effects of stress on startle responsivity would be of interest, particularly with regard to pathologies. In a study with Gulf War veterans suffering from PTSD, enhanced startle responsivity to different kinds of stimuli could be demonstrated, reflecting increased sensitivity to stressful experimental contexts due to fear generalisation (Grillon & Morgan, 1999). It is conceivable that the experienced trauma in PTSD patients leads to a long-term effect based on the shift of amygdala function discussed in section 3.4.2., such that general sensitivity is enhanced, whereas stimulus specific responsivity decreases. Investigating long-term startle responsivity in healthy but stressed participants would provide a measure for comparison with startle responsivity in trauma patients.

In line with other findings (Adolph & Pause, 2012), results of experiments 2 and 3 demonstrated a generally more pronounced startle responsivity to odours than to pictures. As discussed in section 5.6., the startle reflex integrates the input and thus sensory response of the different modalities. Future studies comparing startle responsivity of different modalities,

such as the visual and olfactory system, should therefore always include a startle block with both visual and olfactory stimuli presented together at the same moment for a more reliable comparison. This would also offer the possibility to calculate a delta-value providing an additional measure.

5.10. General conclusion

5.10.1. Concluding "bigger picture"

The organism's stress response is a well-adjusted mechanism initiated from the very onset of the stressful event to cope with the current situation, and ongoing in the aftermath of the stressful experience to consolidate its central information for the future. Hereby, all available sources are concentrated on the main aspects of the situation to make use of any advantage and transfer the pivotal aspects of the experience into long-term memory storage. Thus, the processes during stress cannot be regarded in clear separation of one another, as they all serve the main purpose of an instant and appropriate response to threat and the consolidation of the contents learned from that situation. For this purpose, the available sources support each other, which is why they have to be acknowledged as a holistic mechanism, processing the same main input rather than just separately processing different inputs in a temporally deferred manner. This dissertation therefore aimed at building a bridge between different sensory modalities as well as between encoding and consolidation processes, thus immediate and delayed effects. It was shown that during acute stress, behavioural modifications in terms of fixation measures are detectable. These serve the purpose of focussed attention on and extraction of the central and most crucial aspects of the stressful situation in the very moment of stress for optimal coping. Since it would not be cognitively economical to initiate these processes anew with every time a similar situation reoccurs, other processes come into play to preserve the useful information, maintaining specific attributes of central aspects involved, as well as contextual features. This is by no means restricted to visual input, but concerns all the input available, even if not explicitly and consciously perceived in the stressful situation. Since from the very moment of stress onset, the organism is adjusted to perceive and encode information of no future use rather than to miss out on pivotal information, and rapidly acting processes after encoding further filter the encoded representations and select whatever turned out to be useful. For maximum efficiency, this is an ongoing mechanism preventing crucial information from getting lost, whereas less useful representations become overwritten. This selection is supported by amygdaloidal processes, as the amygdala signals on a moment-to-moment basis. Items initially fixated and encoded might thus not be those items best

remembered in the end, if they in the later course of the situation did not turn out to be essential. However, memory formation is especially promoted from the very beginning of the stressful situation, and memory effects can thus be detected after a short delay (in experiment 3, 40 minutes) as well as after one day, as replicated in experiment 1. The same can be detected for the increased vigilance caused by stress; it leads to pronounced startle responsivity, reflecting enhanced general sensitivity, in the immediate aftermath as well as 24 hours later. Like the neuroendocrine stress response, the startle reflex also is a mechanism to concentrate available sources rapidly in a situation of threat to the organism in a multimodal fashion. Hence, the startle response becomes even potentiated by stress. The findings of the experiments conducted in the framework of this thesis underline the highly specialised and adaptive multi-sensory integration of the stress response and the time course of some main effects caused by it.

5.10.2. Conclusion of the thesis

The current thesis demonstrates modified fixation behaviour under stress, leading to longer fixation times and more fixations in stressed participants. This finding reflects optimised attentional processes promoting encoding of the most relevant information, as it was particularly found for objects central to the situation contrasting peripheral objects. In concert with this effect, memory processes were shown to be enhanced by stress especially for central objects, in direct aftermath as well as one day later. Information bound to the stressor thus promotes increased selective attention to and memory for it, in contrast to features not bound to the stressor and information of a non-stressful episode. This information is not only restricted to visual, but could also be shown for olfactory input. Hereby, formation and consolidation of explicit memory contents is suggested to show a different time-course to implicit memory, dependent on the modality of the encoded stimulus. Whereas explicit memory for visual stimuli seems to increase with time, reflected by more recollection-based memory one day later, explicit memory for an olfactory stimulus seems to decrease, demonstrated by better odour recognition in the immediate aftermath compared to 24 hours later. In contrast, implicit memory for olfactory stimuli seems to benefit from consolidation processes, reflected by a more negative perception of the odour only on the next day. The association of these findings with increased vigilance under stress is nicely shown by generally enhanced startle responsivity under acute stress as well as 24 hours later. Whereas response specificity to the odour in stressed participants seems to decline over the course of one day, it increases for visual stimuli. These findings demonstrate highly adaptive and

specialised processes in response to stress, with modalities operating in concert with each other under acute stress, subsequently revealing separate developments with progressing consolidation processes, in dependence of the salience and intensity of the stimuli in the respective modalities. This thesis thus expands current knowledge about attentional processes in terms of fixation measures under stress, the time course of stress-induced memory effects in two different modalities within one day, and effects of stress on vigilance in terms of startle responsivity, again in two different modalities within one day.

6. References

Abercrombie, H. C., Kalin, N. H., Thurow, M. E., Rosenkranz, M. A., & Davidson, R. J. (2003). Cortisol variation in humans affects memory for emotionally laden and neutral information. *Behavioral Neuroscience*, *117*(3), 505–516.

Abercrombie, H. C., Speck, N. S., & Monticelli, R. M. (2006). Endogenous cortisol elevations are related to memory facilitation only in individuals who are emotionally aroused. *Psychoneuroendocrinology*, *31*(2), 187–196.

Adolph, D., & Pause, B. M. (2012). Different time course of emotion regulation towards odors and pictures: Are odors more potent than pictures? *Biological Psychology*, *91*(1), 65–73.

Ameli, R., Ip, C., & Grillon, C. (2001). Contextual fear-potentiated startle conditioning in humans: replication and extension. *Psychophysiology*, *38*(3), 383–390.

Anderson, A. K., & Phelps, E. A. (2001). Lesions of the human amygdala impair enhanced perception of emotionally salient events. *Nature*, *411*(6835), 305–309.

Anderson, A. K., Wais, P. E., & Gabrieli, J. D. E. (2006). Emotion enhances remembrance of neutral events past. *Proceedings of the National Academy of Sciences*, *103*(5), 1599–1604.

Andres, K. H. (1969). Der olfaktorische Saum der Katze. *Zeitschrift Für Zellforschung Und Mikroskopische Anatomie*, *96*(2), 250–274.

Arnsten, A. F. T. (2009). Stress signalling pathways that impair prefrontal cortex structure and function. *Nature Reviews. Neuroscience*, *10*(6), 410–422.

Atkinson, R.C., Shiffrin, R.M. (1968). Human memory: A proposed system and its control processes. In Spence, K.W., & Spence, J.T. *The psychology of learning and motivation (2).* New York: Academic Press. 89–195.

Baddeley, A. (2001). The concept of episodic memory. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *356*(1413), 1345–1350.

Baddeley, A. (2003). Working memory: looking back and looking forward. *Nature Reviews Neuroscience*, *4*(10), 829–839.

Baddeley, A. (2012). Working Memory: Theories, Models, and Controversies. *Annual Review of Psychology*, *63*(1), 1–29.

Badrick, E., Bobak, M., Britton, A., Kirschbaum, C., Marmot, M., & Kumari, M. (2008). The Relationship between Alcohol Consumption and Cortisol Secretion in an Aging Cohort. *The Journal of Clinical Endocrinology & Metabolism*, *93*(3), 750–757.

Bahrick, L. E., & Lickliter, R. (2014). Learning to Attend Selectively: The Dual Role of Intersensory Redundancy. *Current Directions in Psychological Science*, *23*(6), 414–420.

Balada, F., Blanch, A., & Aluja, A. (2014). Arousal and Habituation Effects (Excitability) on Startle Responses to the International Affective Picture Systems (IAPS). *Journal of Psychophysiology*, *28*(4), 233–241.

Beste, C., Yildiz, A., Meissner, T. W., & Wolf, O. T. (2013). Stress improves task processing

efficiency in dual-tasks. *Behavioural Brain Research*, *252*, 260–265.

Beylin, A. V, & Shors, T. J. (1998). Stress enhances excitatory trace eyeblink conditioning and opposes acquisition of inhibitory conditioning. *Behavioral Neuroscience*, *112*(6), 1327–1338.

Björntorp, P., & Rosmond, R. (2000). Obesity and cortisol. *Nutrition*, *16*(10), 924–936.

Blumenthal, T. D., Cuthbert, B. N., Filion, D. L., Hackley, S., Lipp, O. V., & Van Boxtel, A. (2005). Committee report: Guidelines for human startle eyeblink electromyographic studies. *Psychophysiology*, *42*, 1–15.

Bookbinder, S. H., & Brainerd, C. J. (2016). Emotion and false memory: The context–content paradox. *Psychological Bulletin*, *142*(12), 1315–1351.

Borkenau, P. (1988). The multiple classification of acts and the big five factors of personality. *Journal of Research in Personality*, *22*(3), 337–352.

Borkenau, P., & Ostendorf, F. (1985). *NEO-Fünf-Faktoren Inventar nach Costa und Mccrae - deutsche Fassung* (1.Aufl.). Göttingen: Hogrefe.

Bradley, M. M., Codispoti, M., & Lang, P. J. (2006). A multi-process account of startle modulation during affective perception. *Psychophysiology*, *43*, 486–497.

Bradley, M. M., Codispoti, M., Sabatinelli, D., & Lang, P. J. (2001). Emotion and motivation II: sex differences in picture processing. *Emotion (Washington, D.C.)*, *1*(3), 300–319.

Bradley, M. M., Cuthbert, B. N., & Lang, P. J. (1993). Pictures as prepulse: attention and emotion in startle modification. *Psychophysiology*, *30*(5), 541–545.

Bradley, M. M., Greenwald, M. K., Petry, M. C., & Lang, P. J. (1992). Remembering Pictures: Pleasure and Arousal in Memory. *Journal of Experimental Psychology: Learning, Memory, and Cognition*, *18*(2), 379–390.

Bradley, M. M., Lang, P. J., & Cuthbert, B. N. (1993). Emotion, novelty, and the startle reflex: Habituation in humans. *Behavioral Neuroscience*, *107*(6), 970–980.

Breyer, B. & Bluemke, M. (2016). Deutsche Version der Positive and Negative Affect Schedule PANAS (GESIS Panel). *Zusammenstellung sozialwissenschaftlicher Items und Skalen.*

Buchanan, T. W., Brechtel, A., Sollers, J. J., & Lovallo, W. R. (2001). Exogenous cortisol exerts effects on the startle reflex independent of emotional modulation. *Pharmacology Biochemistry and Behavior*, *68*(2), 203–210.

Bunsey, M., & Eichenbaum, H. (1996). Conservation of hippocampal memory function in rats and humans. *Nature*, *379*(6562), 255–257.

Buschman, T. J., & Miller, E. K. (2007). Top-Down Versus Bottom-Up Control of Attention in the Prefrontal and Posterior Parietal Cortices. *Science*, *315*(5820), 1860–1862.

Butler, W., Ph, D., Jenkins, A., Braff, L., Rausch, J. L., & Geyer, M. A. (1990). Physiological evidence of exaggerated startle response in a subgroup of Vietnam veterans with combat-related PTSD. *American Journal of Psychiatry*, *147*(10), 1308–1312.

Cahill, L., Haier, R. J., Fallon, J., Alkire, M. T., Tang, C., Keator, D., Wu, J., & McGaugh, J.

L. (1996). Amygdala activity at encoding correlated with long-term, free recall of emotional information. *Proceedings of the National Academy of Sciences*, *93*(15), 8016–8021.

Cahill, L., & McGaugh, J. L. (1998). Mechanisms of emotional arousal and lasting declarative memory. *Trends in Neurosciences*, *21*(7), 294–299.

Canli, T., Zhao, Z., Brewer, J., Gabrieli, J. D., & Cahill, L. (2000). Event-related activation in the human amygdala associates with later memory for individual emotional experience. *The Journal of Neuroscience : The Official Journal of the Society for Neuroscience*, *20*(RC99), 1–5.

Cannon, W. B. (1913). The emergency function adrenal medulla in pain and the major emotions. *American Journal of Physiloigy*, *33*, 356–372.

Carmichael, S. T., Clugnet, M.-C., & Price, J. L. (1994). Central olfactory connections in the macaque monkey. *The Journal of Comparative Neurology*, *346*(3), 403–434.

Caulfield, F., Ewing, L., Bank, S., & Rhodes, G. (2016). Judging trustworthiness from faces: Emotion cues modulate trustworthiness judgments in young children. *British Journal of Psychology*, *107*(3), 503–518.

Chajut, E., & Algom, D. (2003). Selective attention improves under stress: Implications for theories of social cognition. *Journal of Personality and Social Psychology*, *85*(2), 231–248.

Christianson, S.-A. (1992). Emotional Stress and Eyewitness Memory: A Critical Review. *Psychological Bulletin*, *112*(2), 284–309.

Christianson, S.-A., Loftus, E. F., Hoffman, H., & Loftus, G. R. (1991). Eye Fixations and Memory for Emotional Events. *Journal of Experimental Psychology: Learning, Memory, and Cognition*, *17*(4), 693–701.

Costa, P. T., & McCrae, R. R. (1992). Normal personality assessment in clinical practice: The NEO Personality Inventory. *Psychological Assessment*, *4*(1), 5–13.

Cowan, N., & Stadler, M. A. (1996). Estimating unconscious processes: Implications of a general class of models. *Journal of Experimental Psychology: General*, *125*(2), 195–200.

Crivelli, C., Jarillo, S., Russell, J. A., & Fernández-Dols, J.-M. (2016). Reading emotions from faces in two indigenous societies. *Journal of Experimental Psychology: General*, *145*(7), 830–843.

Davis, M., & Whalen, P. J. (2001). The amygdala: vigilance and emotion. *Molecular Psychiatry*, *6*(1), 13–34.

Davison, I. G., & Ehlers, M. D. (2011). Neural Circuit Mechanisms for Pattern Detection and Feature Combination in Olfactory Cortex. *Neuron*, *70*(1), 82–94.

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

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

de Kloet, E. R., Oitzl, M. S., & Joëls, M. (1999). Stress and cognition: are corticosteroids good or bad guys? *Trends in Neurosciences*, *22*(10), 422–426.

de Quervain, D. J.-F., Roozendaal, B., & McGaugh, J. L. (1998). Stress and glucocorticoids impair retrieval of long-term spatial memory. *Nature*, *394*(6695), 787–790.

de Quervain, D. J.-F., Roozendaal, B., Nitsch, R. M., McGaugh, J. L., & Hock, C. (2000). Acute cortisone administration impairs retrieval of long-term declarative memory in humans. *Nature Neuroscience*, *3*(4), 313–314.

de Quervain, D. 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.

de Veld, D. M. J., Riksen-Walraven, J. M., & de Weerth, C. (2014). The relation between gaze aversion and cortisol reactivity in middle childhood. *Hormones and Behavior*, *65*(2), 173–178.

Debono, M., Ghobadi, C., Rostami-Hodjegan, A., Huatan, H., Campbell, M. J., Newell-Price, J., Darzy, K., Merke, D. P., Arlt, W., & Ross, R. J. (2009). Modified-Release Hydrocortisone to Provide Circadian Cortisol Profiles. *The Journal of Clinical Endocrinology & Metabolism*, *94*(5), 1548–1554.

Desimone, R., & Duncan, J. (1995). Neural Mechanisms of Selective Visual Attention. *Annual Review of Neuroscience*, *18*(1), 193–222.

Deuter, C. E., Kuehl, L. K., Blumenthal, T. D., Schulz, A., Oitzl, M. S., & Schachinger, H. (2012). Effects of Cold Pressor Stress on the Human Startle Response. *PLoS ONE*, *7*(11), 1–8.

Dickerson, S. S., & Kemeny, M. E. (2004). Acute stressors and cortisol responses: a theoretical integration and synthesis of laboratory research. *Psychological Bulletin*, *130*(3), 355–391.

Ding, X., & Xie, F. (2015). Olfactory Mucosa: Composition, Enzymatic Localization, and Metabolism. In Doty, R. L. (3rd ed.). *Handbook of Olfaction and Gustation*. Hoboken, New Jersey: John Wiley & Sons, Inc., 63-91.

Dolcos, F., LaBar, K. S., & Cabeza, R. (2005). Remembering one year later: Role of the amygdala and the medial temporal lobe memory system in retrieving emotional memories. *Proceedings of the National Academy of Sciences*, *102*(7), 2626–2631.

Doty, R. L. (2001). Olfaction. *Annual Review of Psychology*, *52*(1), 423–452.

Doty, R. L. (2015). *Handbook of Olfaction and Gustation*. (R. L. Doty, Ed.) (Third edit). Hoboken, New Jersey: John Wiley & Sons, Inc.

Dressendörfer, R. A., Kirschbaum, C., Rohde, W., Stahl, F., & Strasburger, C. J. (1992). Synthesis of a cortisol-biotin conjugate and evaluation as a tracer in an immunoassay for salivary cortisol measurement. *The Journal of Steroid Biochemistry and Molecular Biology*, *43*(7), 683–692.

Dusek, J. A., & Eichenbaum, H. (1997). The hippocampus and memory for orderly stimulus relations. *Proceedings of the National Academy of Sciences*, *94*(13), 7109–7114.

Echterhoff, G., & Wolf, O. T. (2012). The stressed eyewitness: the interaction of thematic arousal and post-event stress in memory for central and peripheral event information. *Frontiers in Integrative Neuroscience*, *6*(8), 57(1-12).

Edelmann, R. J., & Neto, F. (1989). Self-reported expression and consequences of embarassment in Portugal and the U.K. *International Journal of Psychology*, *24*, 351–366.

Ekstrand, J. J., Domroese, M. E., Johnson, D. M. G., Feig, S. L., Knodel, S. M., Behan, M., & Haberly, L. B. (2001). A new subdivision of anterior piriform cortex and associated deep nucleus with novel features of interest for olfaction and epilepsy. *The Journal of Comparative Neurology*, *434*(3), 289–307.

Elling, L., Schupp, H., Bayer, J., Bröckelmann, A. K., Steinberg, C., Dobel, C., & Junghofer, M. (2012). The impact of acute psychosocial stress on magnetoencephalographic correlates of emotional attention and exogenous visual attention. *PLoS ONE*, *7*(6), 1-9.

Elzinga, B. M., & Roelofs, K. (2005). Cortisol-Induced Impairments of Working Memory Require Acute Sympathetic Activation. *Behavioral Neuroscience*, *119*(1), 98–103.

Ennis, M., & Holy, T., E. (2015). Anatomy and Neurobiology of the Main and Accessory Olfactory Bulbs. In Doty, R. L. (3rd ed.). *Handbook of Olfaction and Gustation*. Hoboken, New Jersey: John Wiley & Sons, Inc., 157-182.

Fendt, M., Koch, M., & Schnitzler, H.-U. (1994). Amygdaloid noradrenaline is involved in the sensitization of the acoustic startle response in rats. *Pharmacology Biochemistry and Behavior*, *48*(2), 307–314.

Farbman, A. I. (1992). *Cell Biology of Olfaction*. Cambridge University Press, Cambridge.

Frankland, P. W., Scott, B. W., & Yeomans, J. S. (1995). Axons and synapses mediating electrically evoked startle: collision tests and latency analysis. *Brain Research*, *670*(1), 97–111.

Fridlund, A. J., & Cacioppo, J. T. (1986). Guidelines for human electromyographic research. *Psychophysiology*, *23*, 567-589.

Fries, E., Dettenborn, L., & Kirschbaum, C. (2009). The cortisol awakening response (CAR): Facts and future directions. *International Journal of Psychophysiology*, *72*(1), 67–73.

Fuxe, K., Hökfelt, T., & Ungerstedt, U. (1970). Morphological and Functional Aspects of Central Monoamine Neurons. *International Review of Neurobiology*, *13*, 93–126.

Ghio, M., Schulze, P., Suchan, B., & Bellebaum, C. (2016). Neural representations of novel objects associated with olfactory experience. *Behavioural Brain Research*, *308*, 143–151.

Godden, D. R., & Baddeley, A. D. (1975). Context-dependent memory in two natural environments: On land and underwater. *British Journal of Psychology*, *66*(3), 325–331.

Goldstein, D. S. (2003). Catecholamines and stress. *Endocrine Regulations*, *37*(2), 69–80.

Gottfried, J. A. (2009). Function follows form: ecological constraints on odor codes and olfactory percepts. *Current Opinion in Neurobiology*, *19*(4), 422–429.

Gottfried, J. A. (2010). Central mechanisms of odour object perception. *Nature Reviews Neuroscience*, *11*(9), 628–641.

Graf, P., & Schacter, D. L. (1985). Implicit and explicit memory for new associations in normal and amnesic subjects. *Journal of Experimental Psychology: Learning, Memory, and Cognition*, *11*(3), 501–518.

Graham, F. K. (1975). The More or Less Startling Effects of Weak Prestimulation. *Psychophysiology*, *12*(3), 238–248.

Green, D. M., & Swets, J. A. (1966). *Signal detection theory and psychophysics* (1974 reprint with corrections of the original 1966 ed.). Huntington, NY: Robert E. Krieger Publishing Co.

Greenwald, M. K., Bradley, M. M., Cuthbert, B. N., & Lang, P. J. (1998). Startle potentiation: shock sensitization, aversive learning, and affective picture modulation. *Behavioral Neuroscience*, *112*(5), 1069–1079.

Grillon, C., & Morgan, C. A. (1999). Fear-potentiated startle conditioning to explicit and contextual cues in Gulf War veterans with posttraumatic stress disorder. *J. Abnorm. Psychol.*, *108*, 134–142.

Hadley, C. B., & MacKay, D. G. (2006). Does Emotion Help or Hinder Immediate Memory? Arousal Versus Priority-Binding Mechanisms. *Journal of Experimental Psychology: Learning, Memory, and Cognition*, *32*(1), 79–88.

Hamann, S. (2001). Cognitive and neural mechanisms of emotional memory. *Trends in Cognitive Sciences*, *5*(9), 394–400.

Hamann, S. B., Ely, T. D., Grafton, S. T., & Kilts, C. D. (1999). Amygdala activity related to enhanced memory for pleasant and aversive stimuli. *Nature Neuroscience*, *2*(3), 289–93.

Hamm, A.O., Cuthbert, B. N., Globisch, J., & Vaitl, D. (1997). Fear and the startle reflex: Blink modulation and autonomic response patterns in animal and mutilation fearful subjects. *Psychophysiology*, *34*, 97–107.

Hatfield, T., & Gallagher, M. (1995). Taste-potentiated odor conditioning: impairment produced by infusion of an N-methyl-D-aspartate antagonist into basolateral amygdala. *Behavioral Neuroscience*, *109*(4), 663–8.

Heinrichs, N., Hahlweg, K., Fiegenbaum, W., Frank, M., Schroeder, B., & von Witzleben, I. (2002). Validität und Reliabilität der Social Interaction Anxiety Scale (SIAS) und der Social Phobia Scale (SPS). *Verhaltenstherapie*, *12*(1), 26–35.

Heinze, H. J., Mangun, G. R., Burchert, W., Hinrichs, H., Scholz, M., Münte, T. F., Gös, A., Scherg, M., Johannes, S., Hundeshagen, A., Gazaniga, M. S., & Hillyard, S. A. (1994). Combined spatial and temporal imaging of brain activity during visual selective attention in humans. *Nature*, *372*(6506), 543–546.

Hellhammer, D. H., Wüst, S., & Kudielka, B. M. (2009). Salivary cortisol as a biomarker in stress research. *Psychoneuroendocrinology*, *34*(2), 163–171. 6

Henckens, M. J. A. G., Hermans, E. J., Pu, Z., Joels, M., & Fernandez, G. (2009). Stressed Memories: How Acute Stress Affects Memory Formation in Humans. *Journal of Neuroscience*, *29*(32), 10111–10119.

Henderson, J. M. (2007). Regarding scenes. *Current Directions in Psychological Science*, *16*(4), 219–222.

Herman, J. P., Patel, P. D., Akil, H., & Watson, S. J. (1989). Localization and Regulation of Glucocorticoid and Mineralocorticoid Receptor Messenger RNAs in the Hippocampal Formation of the Rat. *Molecular Endocrinology*, *3*(11), 1886–1894.

Hermans, E. J., Henckens, M. J. A. G., Joëls, M., & Fernández, G. (2014). Dynamic adaptation of large-scale brain networks in response to acute stressors. *Trends in Neurosciences*, *37*(6), 304–314.

Herten, N., Otto, T., Adolph, D., Pause, B. M., Kumsta, R., & Wolf, O. T. (2016). Enhanced startle responsivity 24 hours after acute stress exposure. *Behavioral Neuroscience*, *130*(5), 521–530.

Herz, R. S. (1998). Are Odors the Best Cues to Memory? A Cross-Modal Comparison of Associative Memory Stimuli. *Annals of the New York Academy of Sciences*, *855*(1), 670–674.

Herz, R. S., & Cupchik, G. C. (1992). An experimental characterization of odor-evoked memories in humans. *Chemical Senses*, *17*(5), 519–528.

Herz, R. S., & Cupchik, G. C. (1995). The Emotional Distinctiveness of Odor-evoked Memories. *Chemical Senses*, *20*(5), 517–528.

Herz, R. S., Eliassen, J., Beland, S., & Souza, T. (2004). Neuroimaging evidence for the emotional potency of odor-evoked memory. *Neuropsychologia*, *42*(3), 371–378.

Herz, R. S., & Schooler, J. W. (2002). A naturalistic study of autobiographical memories evoked by olfactory and visual cues: testing the Proustian hypothesis. *The American Journal of Psychology*, *115*(1), 21–32.

Het, S., Rohleder, N., Schoofs, D., Kirschbaum, C., & Wolf, O. T. (2009). Neuroendocrine and psychometric evaluation of a placebo version of the "Trier Social Stress Test." *Psychoneuroendocrinology*, *34*(7), 1075–1086.

Het, S., Schoofs, D., Rohleder, N., & Wolf, O. T. (2012). Stress-Induced Cortisol Level Elevations Are Associated With Reduced Negative Affect After Stress: Indications for a Mood-Buffering Cortisol Effect. *Psychosomatic Medicine*, *74*(17), 23–32.

Hinds, J. W., Hinds, P. L., & McNelly, N. A. (1984). An autoradiographic study of the mouse olfactory epitehlium: evidence for long-lived receptors. *Anat. Rec.*, *210*, 375–383.

Hinton, P. B., & Henley, T. B. (1993). Cognitive and affective components of stimuli presented in three modes. *Bulletin of the Psychonomic Society*, *31*(6), 595–598.

Hirst, W., & Phelps, E. A. (2016). Flashbulb Memories. *Current Directions in Psychological Science*, *25*(1), 36–41.

Holbrook, E. H., Wu, E., Curry, W. T., Lin, D. T., & Schwob, J. E. (2011). Immunohistochemical characterization of human olfactory tissue. *The Laryngoscope*, *121*(8), 1687–1701.

Horley, K., Williams, L. M., Gonsalvez, C., & Gordon, E. (2003). Social phobics do not see eye to eye: A visual scanpath study of emotional expression processing. *Anxiety*

Disorders, *17*, 33–44.

Iwai, N., Zhou, Z., Roop, D. R., & Behringer, R. R. (2008). Horizontal Basal Cells Are Multipotent Progenitors in Normal and Injured Adult Olfactory Epithelium. *Stem Cells*, *26*(5), 1298–1306.

Jang, W., Youngentob, S. L., & Schwob, J. E. (2003). Globose basal cells are required for reconstitution of olfactory epithelium after methyl bromide lesion. *The Journal of Comparative Neurology*, *460*(1), 123–140.

Joëls, M., Fernandez, G., & Roozendaal, B. (2011). Stress and emotional memory: A matter of timing. *Trends in Cognitive Sciences*, *15*(6), 280–288.

Joëls, M., Karst, H., DeRijk, R., & de Kloet, E. R. (2008). The coming out of the brain mineralocorticoid receptor. *Trends in Neurosciences*, *31*(1), 1–7.

Joëls, M., Pu, Z., Wiegert, O., Oitzl, M. S., & Krugers, H. J. (2006). Learning under stress: how does it work? *Trends in Cognitive Sciences*, *10*(4), 152–158.

Johnson, B. A., & Leon, M. (2000). Modular representations of odorants in the glomerular layer of the rat olfactory bulb and the effects of stimulus concentration. *The Journal of Comparative Neurology*, *422*(4), 496–509.

Karst, H., Berger, S., Erdmann, G., Schütz, G., & Joëls, M. (2010). Metaplasticity of amygdalar responses to the stress hormone corticosterone. *Proceedings of the National Academy of Sciences*, *107*(32), 14449–14454.

Kastner, S., & Ungerleider, L. G. (2000). Mechanisms of Visual Attention in the Human Cortex. *Annual Review of Neuroscience*, *23*(1), 315–341.

Kensinger, E. A. (2009). Remembering the details: Effects of emotion. *Emotion Review*, *1*(2), 99–113.

Kensinger, E. A., & Corkin, S. (2003). Memory enhancement for emotional words: Are emotional words more vividly remembered than neutral words? *Memory & Cognition*, *31*(8), 1169–1180.

Kevetter, G. A., & Winans, S. S. (1981). Connections of the corticomedial amygdala in the golden hamster. II. Efferents of the "olfactory amygdala" *The Journal of Comparative Neurology*, *197*(1), 99–111.

Kim, J. J., & Diamond, D. M. (2002). The stressed hippocampus, synaptic plasticity and lost memories. *Nature Reviews. Neuroscience*, *3*(6), 453–62.

Kirschbaum, C., & Hellhammer, D. H. (1989). Salivary Cortisol in Psychobiological Research: An Overview. *Neuropsychobiology*, *22*(3), 150–169.

Kirschbaum, C., & Hellhammer, D. H. (1994). Salivary cortisol in psychoneuroendocrine research: recent developments and applications. *Psychoneuroendocrinology*, *19*(4), 313–333.

Kirschbaum, C., Kudielka, B. M., Gaab, J., Schommer, N. C., & Hellhammer, D. H. (1999). Impact of gender, menstrual cycle phase, and oral contraceptives on the activity of the hypothalamic-pituitary-adrenal axis. *Psychosomatic Medicine*, *61*, 154–162.

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.

Kirschbaum, C., Wüst, S., & Strasburger, C. J. (1992). "Normal" cigarette smoking increases free cortisol in habitual smokers. *Life Sciences*, *50*(6), 435–42.

Koch, M. (1999). The neurobiology of startle. *Progress in Neurobiology*, *59*(2), 107–128.

Kocovski, N. L., & Endler, N. S. (2000). Social anxiety, self-regulation, and fear of negative evaluation. *European Journal of Personality*, *14*(4), 347–358.

Kudielka, B. M., Hellhammer, D. H., & Wüst, S. (2009). Why do we respond so differently? Reviewing determinants of human salivary cortisol responses to challenge. *Psychoneuroendocrinology*, *34*(1), 2–18.

Kuhlmann, S., & Wolf, O. T. (2006). Arousal and Cortisol Interact in Modulating Memory Consolidation in Healthy Young Men. *Behavioral Neuroscience*, *120*(1), 217–223.

Laney, C., Campbell, H. V, Heuer, F., & Reisberg, D. (2004). Memory for thematically arousing events. *Memory & Cognition*, *32*(7), 1149–1159.

Lang, P. J., Bradley, M. M., & Cuthbert, B. N. (2008). *International affective picture system (IAPS): Affective ratings of pictures and instruction manual*. Technical Report A-8. University of Florida, Gainesville, FL., 2008.

Lang, P. J., Bradley, M. M., & Cuthbert, B. N. (1990). Emotion, Attention, and the Startle Reflex. *Psychological Review*, *97*(3), 377–395.

Lawrence, J. H., & De Luca, C. J. (1983). Myoelectric signal versus force relationship in different human muscles. *Journal of Applied Physiology: Respiratory, Environmental and Exercise Physiology*, *54*(6), 1653–1659.

LeDoux, J. E. (2000). Emotion Circuits in the Brain. *Annual Review of Neuroscience*, *23*(1), 155–184.

Leung, C. T., Coulombe, P. a, & Reed, R. R. (2007). Contribution of olfactory neural stem cells to tissue maintenance and regeneration. *Nature Neuroscience*, *10*(6), 720–726.

Loftus, E. F., Loftus, G. R., & Messo, J. (1987). Some facts about "weapon focus." *Law and Human Behavior*, *11*(1), 55–62.

Loftus, G. R. (1972). Eye fixations and recognition memory for pictures. *Cognitive Psychology*, *3*(4), 525–551.

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.

Lorig, T. S., Elmes, D. G., Zald, D. H., & Pardo, J. V. (1999). A computer-controlled olfactometer for fMRI and electrophysiological studies of olfaction. *Behavior Research Methods, Instruments, & Computers : A Journal of the Psychonomic Society, Inc*, *31*(2), 370–375.

Lupien, S. J., & McEwen, B. S. (1997). The acute effects of glucocorticoids on cognition:

Integration of animal and human model studies. *Brain Res. Rev.*, *24*(1), 1–27.

Lupien, S. J., Wilkinson, C. W., Brière, S., Ménard, C., Ng Ying Kin, N. M. K., & Nair, N. P. V. (2002). The modulatory effects of corticosteroids on cognition: studies in young human populations. *Psychoneuroendocrinology*, *27*(3), 401–416.

Maass, A., & Köhnken, G. (1989). Eyewitness identification: Simulating the "weapon effect." *Law and Human Behavior*, *13*(4), 397–408.

MacKay, D. G., & Ahmetzanov, M. V. (2005). Emotion, memory, and attention in the taboo stroop paradigm an experimental analogue of flashbulb memories. *Psychological Science*, *16*(1), 25–32.

Mackay, D. G., Shafto, M., Taylor, J. K., Marian, D. E., Abrams, L., & Dyer, J. R. (2004). Relations between emotion, memory, and attention: Evidence from taboo Stroop, lexical decision, and immediate memory tasks. *Memory & Cognition*, *32*(3), 474–488.

MacLennan, R. M. (1993). Interrater Reliability with SPSS for Windows 5 . 0. *The American Statistician*, *47*(4), 292–296.

Mandler, G. (1980). Recognizing: The judgment of previous occurrence. *Psychological Review*, *87*(3), 252–271.

Mather, M. (2007). Emotional Arousal and Memory Binding: An Object-Based Framework. *Perspectives on Psychological Science*, *2*(1), 33–52.

Mather, M., & Sutherland, M. R. (2011). Arousal-Biased Competition in Perception and Memory. *Perspectives on Psychological Science: A Journal of the Association for Psychological Science*, *6*(2), 114–33.

Mattick, R. P., & Clarke, J. C. (1998). Development and validation of measures of social phobia scrutiny fear and social interaction anxiety. *Behaviour Research and Therapy*, *36*(4), 455–470.

Mayeaux, D. J., & Johnston, R. E. (2004). Discrimination of social odors and their locations: role of lateral entorhinal area. *Physiology & Behavior*, *82*(4), 653–662.

McEwen, B. S. (1998). Stress, Adaptation, and Disease: Allostasis and Allostatic Load. *Annals of the New York Academy of Sciences*, *840*(1), 33–44.

McGaugh, J. L. (1966). Time-Dependent Processes in Memory Storage. *Science*, *153*(3742), 1351–1358.

McGaugh, J. L. (2000). Memory - a Century of Consolidation. *Science*, *287*(5451), 248–251.

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.

Merz, C. J., Tabbert, K., Schweckendiek, J., Klucken, T., Vaitl, D., Stark, R., & Wolf, O. T. (2012). Neuronal correlates of extinction learning are modulated by sex hormones. *Social Cognitive and Affective Neuroscience*, *7*(7), 819–830.

Miller, M. W., McKinney, A. E., Kanter, F. S., Korte, K. J., & Lovallo, W. R. (2011). Hydrocortisone Suppression of the Fear-potentiated Startle Response and Posttraumatic Stress Disorder. *Psychoneuroendocrinology*, *36*(7), 970–980.

Mogg, K., & Bradley, B. P. (1998). A cognitive-motivational analysis of anxiety. *Behaviour Research and Therapy*, *36*(9), 809–848.

Mogg, K., McNamara, J., Powys, M., Rawlinson, H., Seiffer, A., & Bradley, B. P. (2000). Selective attention to threat: A test of two cognitive models of anxiety. *Cognition & Emotion*, *14*(3), 375–399.

Moran, J., & Desimone, R. (1985). Selective attention gates visual processing in the extrastriate cortex. *Science*, *229*(4715), 782–784.

Munk, M. H. J., Roelfsema, P. R., Konig, P., Engel, A. K., & Singer, W. (1996). Role of Reticular Activation in the Modulation of Intracortical Synchronization. *Science*, *272*(5259), 271–274.

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

National Academy of Sciences (2014). *Identifying the Culprit: Assessing Eyewitness Identification*. Washington D. C.: National Academies Press.

Nielsen, S. E., Segal, S. K., Worden, I. V., Yim, I. S., & Cahill, L. (2013). Hormonal contraception use alters stress responses and emotional memory. *Biological Psychology*, *92*(2), 257–266.

Öhman, A. (1986). Face the Beast and Fear the Face: Animal and Social Fears as Prototypes for Evolutionary Analyses of Emotion. *Psychophysiology*, *23*(2), 123–145.

Ostendorf, F., & Angleitner, A. (1994). Enthusiasts Contra Pessimists. *Psychological Inquiry*, *5*(2), 159–162.

Packard, P. A., Rodríguez-Fornells, A., Stein, L. M., Nicolás, B., & Fuentemilla, L. (2014). Tracking explicit and implicit long-lasting traces of fearful memories in humans. *Neurobiology of Learning and Memory*, *116*, 96–104.

Paschall, G. Y., & Davis, M. (2002). Olfactory-mediated fear-potentiated startle. *Behavioral Neuroscience*, *116*(1), 4–12.

Pause, B. M., Adolph, D., Prehn-Kristensen, A., & Ferstl, R. (2009). Startle response potentiation to chemosensory anxiety signals in socially anxious individuals. *International Journal of Psychophysiology*, *74*(2), 88–92.

Paylor, R., & Crawley, J. N. (1997). Inbred strain differences in prepulse inhibition of the mouse startle response. *Psychopharmacology*, *132*(2), 169–180.

Payton, C. A., Wilson, D. A., & Wesson, D. W. (2012). Parallel Odor Processing by Two Anatomically Distinct Olfactory Bulb Target Structures. *PLoS ONE*, *7*(4), 1-10.

Peth, J., Vossel, G., & Gamer, M. (2012). Emotional arousal modulates the encoding of crime-related details and corresponding physiological responses in the Concealed Information Test. *Psychophysiology*, *49*(3), 381–390.

Pikkarainen, M., Rönkkö, S., Savander, V., Insausti, R., & Pitkänen, A. (1999). Projections from the lateral, basal, and accessory basal nuclei of the amygdala to the hippocampal formation in rat. *The Journal of Comparative Neurology*, *403*(2), 229–260.

Pilz, P. K. D., Caeser, M., & Ostwald, J. (1988). Comparative threshold studies of the acoustic pinna, jaw and startle reflex in the rat. *Physiology & Behavior*, *43*(4), 411–415.

Plappert, C. F., Pilz, P. K. D., & Schnitzler, H.-U. (1993). Acoustic startle response and habituation in freezing and nonfreezing rats. *Behavioral Neuroscience*, *107*(6), 981–987.

Plessow, F., Kiesel, A., & Kirschbaum, C. (2012). The stressed prefrontal cortex and goal-directed behaviour: Acute psychosocial stress impairs the flexible implementation of task goals. *Experimental Brain Research*, *216*(3), 397–408.

Posner, M. I., & Rothbart, M. K. (2000). Developing Mechanisms of Self-Regulation in Early Life. *Emotion Review*, *3*(2), 207–213.

Prehn, A., Ohrt, A., Sojka, B., Ferstl, R., & Pause, B. M. (2006). Chemosensory anxiety signals augment the startle reflex in humans. *Neuroscience Letters*, *394*(2), 127–130.

Proust, M. (1960). *Swann's way*. (Moncrieff, C. K. S., Trans.). London: Chatto & Windus. (Original work published 1922)

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.

Quas, J. A., Rush, E. B., Yim, I. S., & Nikolayev, M. (2014). Effects of stress on memory in children and adolescents: Testing causal connections. *Memory*, *22*(6), 616–632.

Quas, J. A., Yim, I. S., Edelstein, R. S., Cahill, L., & Rush, E. B. (2010). The role of cortisol reactivity in children's and adults' memory of a prior stressful experience. *Developmental Psychobiology*, *53*(2), 166–174.

Ramos, B. P., & Arnsten, A. F. T. (2007). Adrenergic pharmacology and cognition: Focus on the prefrontal cortex. *Pharmacology & Therapeutics*, *113*(3), 523–536.

Rennaker, R. L., Chen, C.-F. F., Ruyle, A. M., Sloan, A. M., & Wilson, D. A. (2007). Spatial and Temporal Distribution of Odorant-Evoked Activity in the Piriform Cortex. *Journal of Neuroscience*, *27*(7), 1534–1542.

Rensink, R. A. (2000a). The Dynamic Representation of Scenes. *Visual Cognition*, *7*(1–3), 17–42.

Rensink, R. A. (2000b). Visual Search for Change: A Probe into the Nature of Attentional Processing. *Visual Cognition*, *7*(1–3), 345–376.

Rensink, R. A., & Enns, J. T. (1998). Early completion of occluded objects. *Vision Research*, *38*, 2489–2505.

Reul, J. M. H. M., & de Kloet, E. R. (1985). Two Receptor Systems for Corticosterone in Rat Brain: Microdistribution and Differential Occupation. *Endocrinology*, *117*(6), 2505–2511.

Roediger, H. L. (1990). Implicit memory: Retention without remembering. *American Psychologist*, *45*(9), 1043–1056.

Roemer, S., Nees, F., Richter, S., Blumenthal, T. D., & Schächinger, H. (2009). Endogenous cortisol suppression with metyrapone enhances acoustic startle in healthy subjects. *Hormones and Behavior*, *55*(2), 314–318.

Rohleder, N., & Nater, U. M. (2009). Determinants of salivary alpha-amylase in humans and methodological considerations. *Psychoneuroendocrinology*, *34*, 469–485.

Rohleder, N., Nater, U. M., Wolf, J. M., Ehlert, U., & Kirschbaum, C. (2004). Psychosocial stress-induced activation of salivary alpha-amylase: An indicator of sympathetic activity? *Annals of the New York Academy of Sciences*, *1032*, 258–263.

Roozendaal, B. (2000). Glucocorticoids and the regulation of memory consolidation. *Psychoneuroendocrinology*, *25*(3), 213–238.

Roozendaal, B. (2002). Stress and Memory: Opposing Effects of Glucocorticoids on Memory Consolidation and Memory Retrieval,. *Neurobiology of Learning and Memory*, *78*(3), 578–595.

Roozendaal, B. (2003). Systems mediating acute glucocorticoid effects on memory consolidation and retrieval. *Progress in Neuro-Psychopharmacology and Biological Psychiatry*, *27*, 1213–1223.

Roozendaal, B., McEwen, B. S., & Chattarji, S. (2009). Stress, memory and the amygdala. *Nature Reviews Neuroscience*, *10*(6), 423–433.

Roozendaal, B., & McGaugh, J. L. (2011). Memory modulation. *Behavioral Neuroscience*, *125*(6), 797–824.

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

Royet, J. P., Zald, D., Versace, R., Costes, N., Lavenne, F., Koenig, O., & Gervais, R. (2000). Emotional responses to pleasant and unpleasant olfactory, visual, and auditory stimuli: a positron emission tomography study. *The Journal of Neuroscience : The Official Journal of the Society for Neuroscience*, *20*(20), 7752–9.

Rugg, M. D., & Curran, T. (2007). Event-related potentials and recognition memory. *Trends in Cognitive Sciences*, *11*(6), 251–257.

Rugg, M. D., & Yonelinas, A. P. (2003). Human recognition memory: a cognitive neuroscience perspective. *Trends in Cognitive Sciences*, *7*(7), 313–319.

Sänger, J., Bechtold, L., Schoofs, D., Blaszkewicz, M., & Wascher, E. (2014). The influence of acute stress on attention mechanisms and its electrophysiological correlates. *Frontiers in Behavioral Neuroscience*, *8*(10), 1–13.

Schacter, D. L. (1987). Implicit memory: History and current status. *Journal of Experimental Psychology: Learning, Memory, and Cognition*, *13*(3), 501–518.

Schmid, A., Koch, M., & Schnitzler, H.-U. (1995). Conditioned Pleasure Attenuates the Startle Response in Rats. *Neurobiology of Learning and Memory*, *64*(1), 1–3.

Schoofs, D., & Wolf, O. T. (2009). Stress and memory retrieval in women: no strong impairing effect during the luteal phase. *Behavioral Neuroscience*, *123*(3), 547–554.

Schultz, E. W. (1960). Repair of the Olfactory Mucosa with Special Reference to Regeneration of Olfactory Cells (Sensory Neurons). *The American Journal of Pathology*, *37*(1), 1–19.

Schultze, M. (1863). Untersuchungen über den Bau der Nasenschleimhaut, namentlich die Structur und Endigungsweise der Geruchsnerven bei dem Menschen and den Wirbelthieren. *Abh. Naturforsch. Ges.* Halle 7: 1–100.

Schwabe, L., Haddad, L., & Schachinger, H. (2008). HPA axis activation by a socially evaluated cold-pressor test. *Psychoneuroendocrinology*, *33*(6), 890–895.

Schwabe, L., Tegenthoff, M., Hoffken, O., & Wolf, O. T. (2010). Concurrent Glucocorticoid and Noradrenergic Activity Shifts Instrumental Behavior from Goal-Directed to Habitual Control. *Journal of Neuroscience*, *30*(24), 8190–8196.

Schwabe, L., & Wolf, O. T. (2009). The context counts: Congruent learning and testing environments prevent memory retrieval impairment following stress. *Cognitive, Affective, & Behavioral Neuroscience*, *9*(3), 229–236.

Schwabe, L., & Wolf, O. T. (2010). Socially evaluated cold pressor stress after instrumental learning favors habits over goal-directed action. *Psychoneuroendocrinology*, *35*(7), 977–986.

Shackman, A. J., Maxwell, J. S., McMenamin, B. W., Greischar, L. L., & Davidson, R. J. (2011). Stress Potentiates Early and Attenuates Late Stages of Visual Processing. *Journal of Neuroscience*, *31*(3), 1156–1161.

Sharot, T., Delgado, M. R., & Phelps, E. A. (2004). How emotion enhances the feeling of remembering. *Nature Neuroscience*, *7*(12), 1376–1380.

Sharot, T., & Yonelinas, A. P. (2008). Differential time-dependent effects of emotion on recollective experience and memory for contextual information. *Cognition*, *106*(1), 538–547.

Shepherd, G. M. (2007). Perspectives on olfactory processing, conscious perception, and orbitofrontal cortex. *Annals of the New York Academy of Sciences*, *1121*, 87–101.

Shields, G. S., Sazma, M. A., & Yonelinas, A. P. (2016). The effects of acute stress on core executive functions: A meta-analysis and comparison with cortisol. *Neuroscience and Biobehavioral Reviews*, *68*, 651–668.

Shors, T. J., & Mathew, P. R. (1998). NMDA Receptor Antagonism in the Lateral/Basolateral but Not Central Nucleus of the Amygdala Prevents the Induction of Facilitated Learning in Response to Stress. *Learning & Memory*, *5*(3), 220–230.

Silverthorn, D. U. (2009). *Physiology* (4th ed.). München, Germany: Pearson Studium

Skoluda, N., Strahler, J., Schlotz, W., Niederberger, L., Marques, S., Fischer, S., Thoma, M. V., Spoerri, C., Ehlert, U., & Nater, U. M. (2015). Intra-individual psychological and physiological responses to acute laboratory stressors of different intensity. *Psychoneuroendocrinology*, *51*, 227–236.

Smeets, T., Wolf, O. T., Giesbrecht, T., Sijstermans, K., Telgen, S., & Joëls, M. (2009). Stress selectively and lastingly promotes learning of context-related high arousing information. *Psychoneuroendocrinology*, *34*(8), 1152–1161.

Sterling, P., & Eyer, J. (1988). Allostasis: A new paradigm to explain arousal pathology. In Fisher, S., & Reason, J. T. *Handbook of life stress, cognition, and health*. Chicester, NY: Wiley, 629-649.

Stettler, D. D., & Axel, R. (2009). Representations of Odor in the Piriform Cortex. *Neuron*, *63*(6), 854–864.

Stormark, K. M. (2004). Skin conductance and heart-rate responses as indices of covert face recognition in preschool children. *Infant and Child Development*, *13*(5), 423–433.

Strange, B. A., & Dolan, R. J. (2004). ß-Adrenergic modulation of emotional memory-evoked human amygdala and hippocampal responses. *Proceedings of the National Academy of Sciences*, *101*(31), 11454–11458.

Sulmont, C., Issanchou, S., & Köster, E. P. (2002). Selection of Odorants for Memory Tests on the Basis of Familiarity, Perceived Complexity, Pleasantness, Similarity and Identification. *Chemical Senses*, *27*(4), 307–317.

Sutherland, M. R., & Mather, M. (2012). Negative arousal amplifies the effects of saliency in short-term memory. *Emotion*, *12*(6), 1367–1372.

Treisman, A., & Gormican, S. (1988). Feature analysis in early vision: Evidence from search asymmetries. *Psychological Review*, *95*(1), 15–48.

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

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 Ast, V. A., Cornelisse, S., Meeter, M., & Kindt, M. (2014). Cortisol mediates the effects of stress on the contextual dependency of memories. *Psychoneuroendocrinology*, *41*, 97–110.

van Marle, H. J. F., Hermans, E. J., Qin, S., & Fernández, G. (2009). From Specificity to Sensitivity: How Acute Stress Affects Amygdala Processing of Biologically Salient Stimuli. *Biological Psychiatry*, *66*(7), 649–655.

van Steensel, B., van Binnendijk, E. P., Hornsby, C. D., van der Voort, H. T. M., Krozowski, Z. S., de Kloet, E. R., & van Driel, R. (1996). Partial colocalization of glucocorticoid and mineralocorticoid receptors in discrete compartments in nuclei of rat hippocampus neurons. *Journal of Cell Science*, *109*, 787–792.

van Stegeren, A. H., Roozendaal, B., Kindt, M., Wolf, O. T., & Joëls, M. (2010). Interacting noradrenergic and corticosteroid systems shift human brain activation patterns during encoding. *Neurobiology of Learning and Memory*, *93*(1), 56–65.

Vaney, D. I., Sivyer, B., & Taylor, W. R. (2012). Direction selectivity in the retina: symmetry and asymmetry in structure and function. *Nature Reviews Neuroscience*, *13*(3), 194–208.

Ventura-Bort, C., Löw, A., Wendt, J., Moltó, J., Poy, R., Dolcos, F., Hamm, A. O., & Weymar, M. (2016). Binding neutral information to emotional contexts: Brain dynamics of long-term recognition memory. *Cognitive, Affective, & Behavioral Neuroscience*, *16*(2), 234–247.

Vermetten, E., Schmahl, C., Southwick, S. M., & Bremner, J. D. (2007). Positron tomographic emission study of olfactory induced emotional recall in veterans with and without combat-related posttraumatic stress disorder. *Psychopharmacology Bulletin*, *40*(1), 8–30.

Waring, J. D., & Kensinger, E. A. (2011). How emotion leads to selective memory: Neuroimaging evidence. *Neuropsychologia*, *49*(7), 1831–1842.

Watson, D., Clark, L. A., & Tellegen, A. (1988). Development and validation of brief measures of positive and negative affect: the PANAS scales. *Journal of Personality and Social Psychology*, *54*(6), 1063–1070.

Weerda, R., Muehlhan, M., Wolf, O. T., & Thiel, C. M. (2010). Effects of acute psychosocial stress on working memory related brain activity in men. *Human Brain Mapping*, *31*(9), 1418–1429.

Wessel, I., De Kooy, P. V., & Merckelbach, H. (2000). Differential recall of central and peripheral details of emotional slides is not a stable phenomenon. *Memory*, *8*(2), 95–109.

Wiegert, O. (2006). Timing is essential for rapid effects of corticosterone on synaptic potentiation in the mouse hippocampus. *Learning & Memory*, *13*(2), 110–113.

Wiemers, U. S., Sauvage, M. M., Schoofs, D., Hamacher-Dang, T. C., & Wolf, O. T. (2013). What we remember from a stressful episode. *Psychoneuroendocrinology*, *38*, 2268–2277.

Wiemers, U. S., Sauvage, M. M., & Wolf, O. T. (2014). Odors as effective retrieval cues for stressful episodes. *Neurobiology of Learning and Memory*, *112*, 230–236.

Wiemers, U. S., Schoofs, D., & Wolf, O. T. (2012). A friendly version of the Trier Social Stress Test does not activate the HPA axis in healthy men and women. *Stress*, *16*(2), 254–260.

Wiemers, U. S., & Wolf, O. T. (2015). Cortisol broadens memory of a non-stressful social interaction. *Psychopharmacology*, *232*(10), 1727–1733.

Wieser, M. J., Pauli, P., Alpers, G. W., & Mühlberger, A. (2009). Is eye to eye contact really threatening and avoided in social anxiety? - An eye-tracking and psychophysiology study. *Journal of Anxiety Disorders*, *23*(1), 93–103.

Willander, J., & Larsson, M. (2007). Olfaction and emotion: the case of autobiographical memory. *Memory & Cognition*, *35*(7), 1659–1663.

Williams, J. M., Tonymon, P., & Andersen, M. B. (1991). The effects of stressors and coping resources on anxiety and peripheral narrowing. *Journal of Applied Sport Psychology*, *3*(2), 126–141.

Wilson, E., & MacLeod, C. (2003). Contrasting two accounts of anxiety-linked attentional bias: selective attention to varying levels of stimulus threat intensity. *Journal of Abnormal Psychology*, *112*(2), 212–218.

Wilson, D., A., Chapuis, J., & Sullivan, R. M. (2015). Cortical Olfactory Anatomy and Physiology. In Doty, R. L. (3rd ed.) *Handbook of Olfaction and Gustation.* Hoboken, New Jersey: John Wiley & Sons, Inc., 209-223.

Winn-deen, E. S., David, H., Sigier, G., & Chavez, R. (1988). Development of a Direct Assay for Alpha-Amylase. *Clinical Chemistry*, *34*(10), 2005–2008.

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

Wüst, S., Wolf, J., Hellhammer, D. H., Federenko, I., Schommer, N., & Kirschbaum, C. (2000). The cortisol awakening response - normal values and confounds. *Noise & Health*, *2*(7), 79–88.

Xu, W., & Wilson, D. A. (2012). Odor-evoked activity in the mouse lateral entorhinal cortex. *Neuroscience*, *223*, 12–20.

Yehuda, R. (2002). Post-Traumatic Stress Disorder. *New England Journal of Medicine*, *346*(2), 108–114.

Yeomans, J. (2002). Tactile, acoustic and vestibular systems sum to elicit the startle reflex. *Neuroscience & Biobehavioral Reviews*, *26*(1), 1–11.

Yeomans, J. S., & Frankland, P. W. (1996). The acoustic startle reflex: neurons and connections. *Brain Research Reviews*, *21*(3), 301–314.

Yeomans, J. S., & Pollard, B. A. (1993). Amygdala efferents mediating electrically evoked startle-like responses and fear potentiation of acoustic startle. *Behavioral Neuroscience*, *107*(4), 596–610.

Yonelinas, A. P. (2002). The Nature of Recollection and Familiarity: A Review of 30 Years of Research. *Journal of Memory and Language*, *46*(3), 441–517.

Yonelinas, A. P., & Ritchey, M. (2015). The slow forgetting of emotional episodic memories: An emotional binding account. *Trends in Cognitive Sciences*, *19*(5), 259–267.

Young, B. J., & Leaton, R. N. (1994). Fear potentiation of acoustic startle stimulus-evoked heart rate changes in rats. *Behavioral Neuroscience*, *108*(6), 1065–1079.

Young, B. J., Otto, T., Fox, G. D., & Eichenbaum, H. (1997). Memory representation within the parahippocampal region. *The Journal of Neuroscience : The Official Journal of the Society for Neuroscience*, *17*(13), 5183–95.

Zatorre, R. J., Jones-Gotman, M., Evans, A. C., & Meyer, E. (1992). Functional localization and lateralization of human olfactory cortex. *Nature*, *360*(6402), 339–340.

Zippel, H. P. (1993). Historical aspects of research on the vertebrate olfactory system. *Naturwissenschaften*, *80*(2), 65–76.

7. Appendix

7.1. Curriculum vitae

Nadja Herten

***12 August 1980, Aachen, Germany**

Department of Cognitive Psychology
Ruhr University Bochum
GAFO 02/384
Universitätsstraße 150
44780 Bochum

Nadja.Herten@rub.de

Education

Gymnasium	
1991 – 1999	Rhein-Maas-Gymnasium, Aachen, Germany
1999 – 2001	Viktoriaschool, Catholic Gymnasium, Aachen **Degree: Abitur**
Apprenticeship	
07/2005 - 07/2008	Mechatronics Engineer Mies-van-der-Rohe-School, Aachen
University	
10/2008 - 09/2011	Psychology, Ruhr University Bochum, Germany **Degree: Bachelor of Science Psychology** Topic: The correlation between contingency detection and socio-emotional conspicuity in 6 months old infants.

10/2011 -	Cognitive Neuroscience, Ruhr University Bochum
09/2013	**Degree: Master of Science in Cognitive Neuroscience**
	Topic: The correlation between visual-proprioceptive and visual-tactile contingency detection in infants of 6 and 9 months of age.
10/2013 -	Cognitive Psychology, International Graduate School of
now	Neuroscience, Research School of the Ruhr University Bochum
work experience	
10/2013 -	Department of Cognitive Psychology,
now	Ruhr University Bochum
Teaching	
WS 13/14	B Sc Psychology, Introduction to empirical research work
SS 14	B Sc Psychology, Cognition II seminar course
WS 14/15	B Sc Psychology, Cognition I seminar course
SS 15	B Sc Psychology, Cognition II seminar course
Tutoring	
SS 12	B Sc Psychology, “The human brain - a painting- and craftsmanship course”
	Psychological Internships
08/2009 -	Official Prison Visitor at the correctional facility, Bochum
01/2010	chair in Criminology at Ruhr University Bochum
08/2010 -	Professional traineeship at the practice for psychotherapy
11/2010	Detlef W. Timp, Gelsenkirchen, Germany
11/2012 -	Research internship, Department of Applied Developmental
08/2013	Psychology, Ruhr University Bochum

Engineering Internships

02/2003 -	Measurement and Test Engineering,
09/2003	Coskun-Engineering, Special Purpose engineering, Aachen
07/2004 -	Professional practical training Mechatronics,
12/2004	Coskun-Engineering, Aachen

Bochum, 16 January 2017

1. List of publications

Peer reviewed articles:

Herten, N., Otto, T., Adolph, D., Pause, B. M., Kumsta, R., & Wolf, O. T. (2016). Enhanced startle responsivity 24 hours after acute stress exposure. *Behavioral Neuroscience, 130*(5), 521–530. http://doi.org/10.1037/bne0000156

Herten, N., Otto, T., & Wolf, O. T. (2017). The role of eye fixation in memory enhancement under stress – An eye tracking study. *Neurobiology of Learning and Memory, 140*(2017), 134-144. http://dx.doi.org/10.1016/j.nlm.2017.02.016

Herten, N., Pomrehn, D., & Wolf, O. T. (2017). Memory for objects and startle responsivity in the immediate aftermath of exposure to the Trier Social Stress Test. *Behavioural Brain Research, 326*(2017), 272-280. http://dx.doi.org/10.1016/j.bbr.2017.03.002

Poster presentation:

Herten, N. & Wolf, O. T. (2015). *Der Einfluss von Stress auf die menschliche Startle Reaktion*. 41. Tagung "Psychologie und Gehirn", Frankfurt am Main, Germany.

Conference talk:

Herten, N., Pomrehn, D., & Wolf, O. T. (2015). *The sound of stress: Was die Sprachgrundfrequenz (F0) über das Stressniveau des Sprechers aussagt.* Symposium „Der Stress in der Stimme – Stimmgrundfrequenz als nützlicher Indikator für emotionale Erregung in Partnerschaft und Psychotherapie?!" by Einsle & Hahlweg, 9. Workshopkongress für Klinische Psychologie und Psychotherapie, Dresden, Germany.