Stress, emotion and cognition : role of mineralo- and glucocorticoid receptors

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Chapter 7

General discussion

Outline

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The objective of this thesis was to identify the distinct contribution of corticosteroids and their receptors to the integration of emotional and cognitive processes. I focussed on the following questions:

1. Do corticosteroids affect emotion and cognition via differential MR and GR **activation**? Are emotion and cognition correlated?
2. Do emotion and cognition correspond to distinct MR, GR **expression** and stress susceptibility as it is expressed in two mouse strains?
3. Can strain differences in emotion and cognition for a negative event be eliminated by manipulating endogenous corticosterone levels?
4. Does the time of treatment (before or directly after the negative event) differentially influence memory formation and extinction?
5. What is the specific function of MR during memory formation and extinction of a stressful emotional experience?

Below I will discuss the results of this thesis by addressing these main questions **(sections 7.1** to **7.5)**, propose a model that describes the integration between the glucocorticoid stress system, emotion and cognition **(section 7.6)**, and address the implications for the development and possible treatment of stress-related diseases like post-traumatic stress disorder (PTSD, **section 7.7**). I will give perspectives for future research **(section 7.8)** and finalize with the general conclusions **(section 7.9)**.

7.1 Do corticosteroids affect emotion and cognition via differential MR and GR activation? Are emotion and cognition correlated?

In **chapter 2**, the correlation between emotion and cognition, and the influence of differential MR and GR activation on this correlation are examined. Combined emotional and cognitive testing was performed in a positively stimulated spatial task using C57BL/6J mice with distinct MR and GR activation. Results show that emotion and cognition are indeed correlated. With the help of principal component analysis I demonstrate that anxiety and motivation are correlated to learning, and that both emotions are especially important during the early phase of learning.

In addition, the results show that distinct MR and GR activation differentially affects emotional and cognitive processes. When confronted with a novel situation, continuous predominant MR activation is beneficial for the emotional state. This state is expressed by low anxiety, high motivation and high directed exploration, which allows to gain detailed knowledge of the environment. Remarkably, this condition of predominant MR activation does not result in better learning and memory. To gain profit from this adaptive behaviour when

confronted with the same situation again, additional consolidation is required. This could be achieved by the concurrent activation of the GR [1-4]. Indeed, mice with continuous predominant MR and additional moderate GR activation are fast learners. They display low anxiety and arousal together with high directed explorative behaviour as well as improvement of cognitive performance. Thus, moderate GR activation contributes to the facilitation of memory. Further increase to continuous GR activation, however, induces strong emotional arousal at the expense of cognitive performance. This has also been found in previous research [5-7], however the present study has the advantage of combining such emotional and cognitive effects into a clear correlation.
Several studies have addressed the issue that increasing corticosterone levels affect cognitive functioning in complex spatial tasks in a dose-dependent inverted-U-shaped fashion, with MR and GR as molecular candidates for this effect [8-12]. The results of this thesis add novel information to the inverted U-shaped function of corticosterone, by demonstrating for the first time how the integration of corresponding emotional parameters affects cognitive processes of learning and memory.

7.2 Do emotion and cognition correspond to distinct MR, GR expression and stress susceptibility as it is expressed in two mouse strains?

To answer this question, glucocorticoid stress system markers together with emotional expression, learning and memory were studied in two distinct mouse strains **(chapter 3)**. Results indeed show corresponding MR, GR expression, stress susceptibility, emotion and cognition in BALB/c and C57BL/6J mice. Lower hippocampal MR and GR mRNA expression, but elevated GR mRNA in prefrontal cortex and GR protein in the amygdala of BALB/c mice coincides with increased stress susceptibility, high emotional expression and superior cognitive performance in a spatial test. High hippocampal MR and GR mRNA expression and high GR protein in hippocampus of C57BL/6J mice corresponds with less stress susceptibility and inferior cognitive performance. The latter is stimulus-response driven and lacks emotional contribution.
This data corresponds to literature which describes that similar differences in MR and GR expression coincide with distinct stress dependent neuroendocrine regulation [13-16], emotion [17-21] and cognition [1;3;17].
However, it adds novel insights on how genetic variation of the glucocorticoid stress system could affect the correlation between emotion and cognition.

In summary, **chapters 2** and **3** show a clear contribution of the glucocorticoid stress system acting via MR and GR on the integration of emotion and cognition; **chapter 2** shows that moderate levels of corticosterone coincide with

optimal emotional state and cognitive performance, and **chapter 3** shows that in highly stress sensitive mice emotions positively contribute to optimal cognitive performance. MR and GR may play a coordinating role for these emotional and cognitive processes [2;22].

7.3 Can strain differences in emotion and cognition for a negative event be eliminated by manipulating endogenous corticosterone levels?

In **chapter 4,** the development of fear behaviour and the expression of fear memories are examined in naive BALB/c and C57BL/6J mice. A paradigm was designed in which several aspects of possible strain dependent fear responses could be tested. First, the developed setup allows assessing both context- and cue-related fear behaviour in one experimental procedure. This enables the detection of generalized and specific fear responses. Second, applying in depth behavioural observation allows to differentiate between qualities of fear behaviour. While scanning expresses active fear behaviour, freezing indicates passive, more intense fear expression. It was expected that due to their distinct stress susceptibility, emotional expression and cognitive functioning described in **chapter 3,** learning and memory of fearful events would also differ.

Chapter 4 supports the findings of **chapter 3,** demonstrating that BALB/c mice are the more stress susceptible strain displaying twofold higher corticosterone levels after fear conditioning and fear memory testing than C57BL/6J mice. In addition, a clear strain dependent (i) expression of fear behaviour by scanning and freezing and (ii) differentiation between context and cue related fear is observed. BALB/c mice display higher freezing than scanning behaviour during acquisition and memory testing, while C57BL/6J mice show more scanning than freezing behaviour. This reflects high passive coping behaviour in BALB/c mice and increased active coping behaviour in C57BL/6J mice. The latter has been suggested to reflect escape behaviour in expectance of the aversive event [23]. Interestingly, MR expression in these strains could contribute to the distinct coping behaviour: less MR function, as observed in BALB/c mice, appears to facilitate fear induced freezing [17;24].

BALB/c and C57BL/6J mice also display different acquisition of fear behaviour and fear memory. BALB/c mice display high levels of extreme fear (freezing) during context episodes of the acquisition compared to C57BL/6J mice. Strain differences are also present during fear memory testing: C57BL/6J mice very quickly change their fear behaviour between context and cue episodes, showing low freezing during context and high freezing when the cue is switched on. In contrast, BALB/c mice display a generalized high fear response independent of context or cue episodes. This stimulus driven cognitive performance of C57BL/6J mice and strong contribution of spatial (contextual) stimuli in BALB/c mice

reflects the strain specific cognitive performance described in **chapter 3.** Thus, the cognitive performance keeps the strain-specific characteristics independent of the motivational aspects of the task (adverse for fear conditioning; appetitive for the hole board)
Also here, distinct expression of MR and GR in the brain (hippocampus for context and amygdala for specific stimuli [25]), and stress susceptibility could underline strain dependent cognitive performance. Furthermore, the data presented emphasize the distinct emotional and cognitive functioning of these mice.

The results presented in **chapter 4** lead to the question whether the distinct stress susceptibility of BALB/c and C57BL/6J mice as expressed by different endogenous corticosterone levels, would underlie the strain specific emotion and cognition for a negative event. Thus will changing endogenous corticosterone levels either potentiate or diminish their distinct fear behaviour and fear memory? Since corticosterone effects are known to be facilitating as well as impairing for memory formation and extinction [2;26;27], therapeutic effects of the hormone might be unveiled. To address this question, corticosterone was given to BALB/c and C57BL/6J mice before or after acquisition **(chapter 5)**.
Interestingly, during this follow-up experiment **(chapter 5)** it appears that the kind of fear conditioning apparatus specifically affects the behaviour of C57BL/6J mice during the conditioning phase. Experiments presented in **chapter 4** used a transparent box, while a black, non-transparant box was used **chapter 5.** C57BL/6J mice showed less freezing during context episodes in the transparent box **(chapter 4)**. This finding is in line with the stimulus-driven behaviour of this mouse strain. However, the difference in expression of freezing as fear behaviour is bound to the acquisition phase, as naïve mice of both strains do display similar strain specific memory in both experiments **(chapters 4 and 5)**. BALB/c mice show generalized strong fear memory, while C57BL/6J mice clearly discriminate alternating context-and cue episodes.
Remarkably, corticosterone treatment strengthens the strain-dependent fear behaviour. The existing strong distinction between context- and cue-related fear in C57BL/6J mice becomes even more prominent. In BALB/c mice, corticosterone destabilizes fear memory to the benefit of facilitated extinction. It seems unlikely, that further increasing the dose of corticosterone in C57BL/6J mice would result in processing of fear comparable to the high stress sensitive, high corticosterone secreting BALB/c mouse.

In summary, data from **chapter 3** clearly shows a strain specific anxiety-like behaviour in novel environments which could be the consequence of distinct

MR and GR expression and stress susceptibility of BALB/c and C57BL/6J mice **(chapter 4)**. Modulation of endogenous corticosterone levels does not eliminate the strain specific fear behaviour **(chapter 5)**.

7.4 Does the time of treatment (before or directly after the negative event) differentially influence memory formation and extinction?

Besides determining the possible diminishing or potentiating effect of corticosterone treatment on the strain specific fear memory, the experiment discussed in **chapter 5** also reveals the influence of timing of corticosteroid action on cognition [3;12;26;28;29]. It is expected that corticosterone treatment before training (pre-acquisition) influences behaviour during acquisition *and* consolidation, while treatment after the aversive event (i.e., the fear conditioning procedure; post-acquisition) will affect solely memory consolidation.
Indeed, **chapter 5** shows that timing of corticosterone treatment does influence fear memory differently, with strain-dependent characteristics. For BALB/c mice, corticosterone treatment *before* acquisition hardly affects fear memory, while corticosterone treatment *after* acquisition apparently destabilizes consolidation and thereby facilitates extinction. In C57BL/6J mice, corticosterone treatment *before* the acquisition results in increased fear memory and impaired extinction of cue related fear, while corticosterone treatment *after* acquisition does not clearly affect fear memory. The presence of additional corticosterone during acquisition has opposite effects on fear memory. This is a novel and unexpected result. The general idea is that corticosterone facilitates fear memory consolidation, when given in *context* with the fear conditioning (pre- and post-acquisition). The present fear conditioning paradigm allowed to demonstrate the profound differences of pre- and post-acquisition treatment of corticosterone. On top of that, the effect of corticosterone is strain-dependent. It will be a challenge to unravel the underlying molecular mechanisms. At present, I may speculate that these time-of-treatment and strain-dependent effects corticosterone point to fast non-genomic actions of corticosterone mediated by membrane located low affinity MR [30].

In summary, as observed in **chapters 2** and **3**, **chapters 4** and **5** demonstrate distinct strain-dependent corticosterone levels and other markers of a differentially regulated glucocorticoid stress system, as well as behavioural patterns to spatial (context) and more specific stimuli (cue). Emotional expressions and memory performance show large individual differences. Distinct MR and GR expression in the brain areas specific for these memory processes could be contributing to the strain dependent memory processes **(section 7.6)**.

Additional exogenous corticosterone treatment influences memory for the adverse emotional event depending on time of administration (i.e. either before or after acquisition) and mouse strain. We conclude that genetic background and time of corticosterone action during processing of stressful information are modifiers of fear memory with interesting translational implications for anxiety-related diseases. How these results can be used in the translational research involving modelling and treating stress-related diseases such as PTSD will be discussed in **section 7.7**.

7.5 What is the specific function of MR during memory formation and extinction of a stressful emotional experience?

Previous research has shown that corticosterone action via MR influences behavioural reactivity and possibly also cognitive functioning [2;17;31-35]. To determine the specific contribution of MR to these behaviours, (female) mice with ablated MR in the forebrain ($MR^{CaMKCre}$ mice [17]) were tested for behavioural responses towards novelty and cognitive processing **(chapter 6)**.
$MR^{CaMKCre}$ mice show higher arousal and less locomotion in a novel environment compared to control mice, although only when the $MR^{CaMKCre}$ mice were pre-stressed. This increase in passive coping corresponds to previous findings using these mice [17], and to other experiments using mice with less MR function [24]. It appears that absence of MR function over time changes stress-induced behaviour. In addition, the timeframe of this behavioural effect could imply the involvement of fast-non genomic corticosterone actions via membrane MR in control mice [30].
Results also show that conditioned behaviour is affected by the absence of forebrain MR function. $MR^{CaMKCre}$ mice display enhanced cue-related fear behaviour during acquisition and persistently increased fear memory for the context. Besides the MR mediated effect, the importance of GR contribution to conditioned behaviour has to be considered as well [20;21], especially since these $MR^{CaMKCre}$ mice have higher GR expression compared to controls.
Chapter 6 presents very relevant data on the contribution of the MR to anxiety-like behaviour and the fear-related learning and memory processes with a high potential for translational research **(section 7.7).** In contrast, animal models in which the MR is overexpressed in the brain should be considered with caution due to the use of different promoters and MR expression in areas where normally no MR expression is found [18;32;33].
Interestingly, fear responses during acquisition and memory tests of the female control mice for the $MR^{CaMKCre}$ ablation are rather comparable to the fear responses observed in naïve male C57BL/6J mice in **chapters 4** and **5**. Both female control mice and male C57BL/6J mice discriminate between context- and

cue-related fear responses. This finding is relevant, since the genetic background of the $MR^{CaMKCre}$ control mice is predominantly C57BL/6J. It appears that the similarity in background has more influence on fear related behavioural response than a possible effect of gender.

In conclusion, **chapter 6** clearly specifies which unconditioned behaviours are under modulating influence of MR and how disrupted MR function influences different stages of learning and memory. Since the balance of MR/GR is shifted towards a larger contribution of GR-mediated effects, these results furthermore stress the importance of coordinated glucocorticoid receptor actions [2;22].

7.6 Proposed model of integrating glucocorticoid stress system, emotion and cognition

This thesis presents several experiments addressing the interaction between the glucocorticoid stress system with emotion and cognition. The experiments focus on pharmacological activation of MR and GR, naturally occurring variances in MR and GR expression and genetic modification of MR. To gain insight in the complete behavioural spectrum, both low and high emotional behavioural test conditions are used [25;36-39] .

The obtained results lead to several conclusions (see **section 7.9**), but the following two conclusions are relevant for the proposed model:

1. Emotion and cognition interact strongly.
2. Expression and activation of the corticosteroid receptors MR and GR clearly influence the contribution of emotional components to cognitive functioning.

How emotion and cognition interact and how the glucocorticoid stress system influences this interaction is presented in the proposed model in figure 1. BALB/c and C57BL/6J mice, with distinct MR and GR expression in the hippocampus, amygdala and PFC already exhibit different behaviour during unstressed, non-activated conditions. Indeed, the experiments performed in this thesis support this. The behavioural pattern of the mice in unstressed conditions, their cognitive performance and response to acute corticosterone treatment will allow to use both strains to model different aspects of stress-related disorders, like PTSD and generalized anxiety disorder **(section 7.5)**. For reasons of clarity, BALB/c and C57BL/6J mice are addressed separately in the proposed model.

In naive BALB/c mice, emotions contribute strongly to unconditioned and conditioned behaviour. This large emotional component parallels and could even directly enhance cognitive performance in this mouse strain. The learning strategy of BALB/c mice depends on complex (spatial) stimuli and is well orchestrated. When introducing a stressor or corticosterone treatment, the emotional component increases and cognitive performance becomes impaired. In naive C57BL/6J mice, the contribution of emotions to unconditioned and conditioned behaviour is much less expressed and no clear interaction is observed*. Their learning strategy focuses on the processing of specific stimuli, and performance in complex, spatial tasks is inferior compared to BALB/c mice. When introducing a stressor or injecting additional corticosterone, the emotional component becomes stronger, accompanied by facilitation in cognitive functioning.

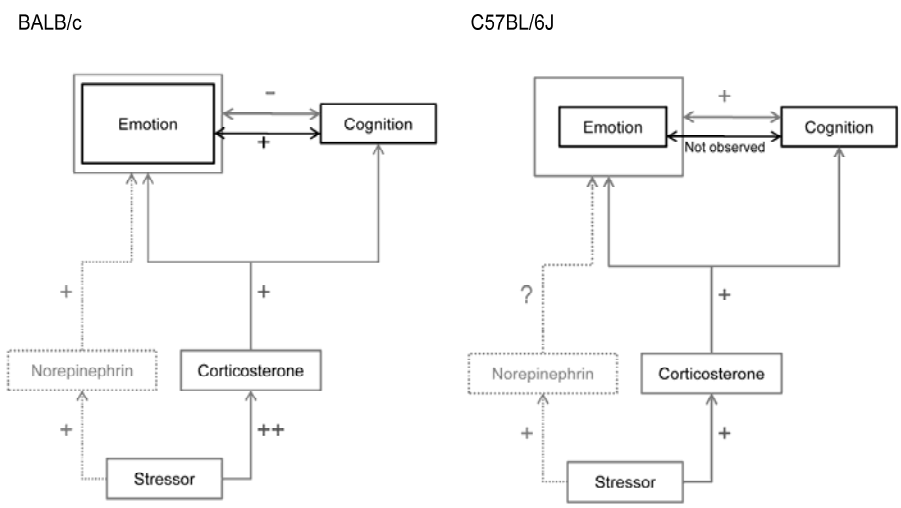

Figure 1. Schematic view of proposed model that describes the interaction between the glucocorticoid stress system, emotion and cognition. **Unstressed conditions**: the black connection line between the emotion and cognition box indicates the relationship between emotion and cognition in naive BALB/c and C57BL/6J mice. **The effect of a stressor or corticosterone**: the grey connection line and boxes indicate the effect of a stressor or corticosterone injection + (facilitation) and – (impairment) on emotion and cognitive functioning. This thesis does not experimentally address the response of the sympathetic nervous system to stress. However, due to a possible contribution of the noradrenergic system to the behaviour of BALB/c mice (see discussion **chapter 5**), this is included in the proposed model (dotted light grey boxes and lines).

*Note: **Chapter 2** does describe a clear interaction between emotion and cognition in C57BL/6J mice. However, most of these mice have surgically removed adrenals and are subjected to continuous corticosterone exposure via implanted pellets. The absence of the endogenous glucocorticoid stress response makes this ADX (mouse) model appropriate to determine the effect of differential MR and GR activation on interacting emotion and cognition. The C57BL/6J mice in this model are not considered as naive C57BL/6J mice depicted in figure 1. Sham operated mice with an intact adrenocortical (glucocorticoid) stress and adrenal medullary response differ significantly from the surgically manipulated mice in the factor describing the interaction between emotion and cognition.

7.7 Translational approach: from mouse to man

PTSD is a well known stress related disease characterised by disrupted glucocorticoid stress system regulation and altered cognition for the emotional event [40;41]. One of the major behavioural symptoms is the intrusive uncontrollable reoccurrence of traumatic memory [42].
This thesis describes several experiments that study the relation of the glucocorticoid stress system and memories for an adverse, most likely traumatic event **(chapters 4, 5 and 6)**. Results obtained with these experiments may provide information on how fear memories develop and perhaps even point out new possibilities for the therapy of pathological fear memories. First, I will discuss the translational value of the models presented in this thesis, followed by a comparison with existing models.

Translational value of presented models

It is of relevance to note that patients suffering from PTSD often display symptoms in the presence or imagination of particular stimuli [42], whereas in generalized anxiety disorder, behavioural and emotional reactions often emerge in the absence of a particular stimulus, or are interconnected with a more complex environment [43]. BALB/c and C57BL/6J mice were used to determine to what extend differences in genetic background related to the glucocorticoid stress system contribute to the formation and extinction of fear memories. We carefully studied the separate phases of acquisition, consolidation, retrieval and extinction of fear to complex (*context:* the environment) and simple stimuli (*cues*: light and tone). The highly emotional and stress sensitive BALB/c mice acquire fear and remember fear-related conditions differently from the less emotional and less stress sensitive C57BL/6J mice. With respect to "PTSD like" symptomatology, naive C57BL/6J mice seem to be more vulnerable to cue-specific fear memories, easily to trigger and most likely expressing "flashback" memories. Naive BALB/c mice present more the phenotype with higher anxiety-related behaviour and generalization of fear memory to discrete stimuli and context.

Using glucocorticoids as therapeutics to restrict the consolidation and facilitate the extinction of fear [44;45], BALB/c mice treated with corticosterone appear to be a good model. BALB/c mice respond with alleviated fear memory preferentially when treated *after* the adverse experience. Similar results have been described in PTSD patients after glucocorticoid treatment [45;46]. On the other hand, BALB/c mice do not show the cue-related specificity of fear memories that is characteristic for PTSD. This is characteristic for C57BL/6J mice that show stimulus specific fear memories. Based on results of C57BL/6J mice, I may predict that increased levels of glucocorticoids present during an adverse, stressful event will be a risk factor for PTSD. C57BL/6J mice present an animal model for studying the strong acquisition and consolidation of fear memories for a specific stimulus as seen in PTSD patients.
The strain-specific effects of corticosterone on fear memories highlight the relevance of the genetic background related to the glucocorticoid stress system for therapeutic efficacy.

Comparison with existing models

Many studies described in literature focus on modelling PTSD related fear memory in rodents using fear conditioning paradigms. I will compare some of these models with the ones presented in this thesis, focussing on glucocorticoid stress system activation/ modulation and genetic variance.
A common approach to acquire rodent models to study PTSD related fear involves (repeated) exposure to a stressor or corticosteroid treatment. Fear conditioning uses the same approach since it involves the acquisition of fear memory due to the repeated exposure to stressful events (shocks). However, several studies have shown that additional single or repeated exposures to a stressor enhances fear memory [47-50], possibly due to a GR-mediated effect [51]. The model described in this thesis (treating C57BL/6J mice with corticosterone before acquisition) agrees with these models. In addition, the present paradigm allows a more specific analysis of cognitive processes such as the contribution of context and cue to fear memory formation and extinction. These models are very valuable for translational research as they allow to study how environmental stressors can contribute to the formation of fear memories in humans.
Besides facilitating effects of stressors and corticosteroids on the formation of fear memory, and thus PTSD symptomatology, corticosteroid treatment in rodents can also be used to model possible intervention of established fear memories. This has been shown in **chapter 5** using BALB/c mice, but also by the proposed model of Cai and colleagues (2006). In their model, treatment with corticosterone after reactivation of established fear impairs later recall of that

fear memory [52], and even could improve extinction [26]. Others elaborate on this, showing with their models that the corticosteroid effect depends on the strength of fear memory [53;54] .
As glucocorticoid treatment in recent human studies confirmed [44;45], these rodent models are well suited to further study the mechanisms of traumatic fear memories.
Other rodent models focus on the genetic factors that underlie development and disruption of fear memory. Some of these models propose transcription factors or amygdala functioning [55;56]. However only few rodent models include genetic modulation of glucocorticoid associated genes. Chourbaji and colleagues propose that mice with GR overexpression might be suitable as model for increased fear memory, arguing that increased GR activation contributes to strengthening of consolidation of fear memories [57]. **Chapter 6** of this thesis describes how MR ablation can model the impairment in adjusting fear responses to a safe situation, as observed in PTSD patients. Also in this MR dysfunctional mice, increased GR activation might contribute to the strong fear-memories, resistant to extinction. Thus, mouse models focussing on the glucocorticoid stress system might indeed be very helpful in determining which genes are involved in the establishment and disruption of fear memories.
Another option is to use animal models that rely on naturally occurring genetic variance. Some of them involve the use of different inbred mouse strains [58] or rely on the crossbreeding of strains [59;60]. The experiments in this thesis demonstrate that C57BL/6J and BALB/c mice are very suitable in determining how naturally occurring genetic differences in the glucocorticoid stress system correlate with anxiety-related behaviour, fear memories and other cognitive abilities.

7.8 Perspectives

The chapters in his thesis present new insights on how the glucocorticoid stress system affects the integration between emotion and cognition. Knowledge on this interaction is sparse, but very much needed when addressing vulnerability and treatment of stress related diseases such as depression and PTSD. The tools developed here include animal models, detailed behavioural and statistical analysis, and can be used to further study various aspect of the integration between the glucocorticoid stress system, emotion and cognition. Below, I will present several ideas for future experiments regarding the (i) underlying mechanisms of glucocortiocoid stress system interaction on emotion and cognition and (ii) translational research.

Mechanisms of glucocorticoid action

An interesting line of research is to determine in more detail how and which of the brain structures involved in the glucocorticoid stress system modulate the effects on emotion and cognition. It would be very interesting to study the contribution of amygdala versus hippocampus in BALB/c and C57BL/6J mouse strains. This can be achieved by fMRI studies using mice with distinct glucocorticoid stress system activation due to knockout or pharmacological modulation tested in either positively or negatively stimulated cognitive tasks.

Additional to these experiments, mouse studies regarding the smaller hippocampal volume after traumatic events in humans should be performed. Such studies, would give more insight in the question whether smaller hippocampal volume reflects higher vulnerability to strong fear related memory formation or if less volume is a result of experiencing a negative event. Interestingly, Penet and colleagues have found a relative small hippocampus in C57BL/6J mice [61]. So this strain, possibly in combination with others, would be very suitable for such research.

Another very promising tool is the use of siRNA to specifically knockdown a gene of interest in a very limited spatial domain. This means that MR and GR function can be determined in specific sub-areas of the hippocampus, amygdala and PFC, and that therefore their contribution to strain specific behaviour can be elucidated. Experiments performed in collaboration with L. van Hooijdonk, E. Vreugdenhil, C. Fitzsimons and colleagues have already revealed first results, showing that the GR in the DG of the hippocampus affects context and cue-related fear memories and extinction processes (unpublished data).

In addition to this, further experiments using inducible ablation of GR and MR should be performed. A start has already been made in **chapter 6**, using mice with MR ablation. However, a similar study on the specific role of limbic GR would complement this data. BALB/c and C57BL/6J mice are very suitable for such research as it appears that these strains have distinct GR protein **(chapter 3)**.

When addressing MR function, non-genomic actions of glucocorticoids via membrane bound MR should not be overlooked [30]. This thesis suggests that it is the fast non-genomic MR mediated action of glucocorticoids that affects behaviour during acquisition of fear memory. Which emotional and cognitive processes are under influence of these effects is yet unknown.

Translational research

One of the next steps concerning translational research is to study fear related cognitive processes and their modification over an extended time interval. The delay between the traumatic event and recurrence of fear memory in PTSD

patients can be up to months or even years [62;63]. In this thesis, relatively short time spans (days) of occurrence and extinction of fear memories were used.
Another promising observation described in this thesis is the influence of disrupted MR function on formation of fear memory and its extinction. Ablation of MR in the forebrain increases fear memory, but also impairs adaptation to the relative safe situation, i.e. when the cue is not followed by shock anymore. These $MR^{CaMKCre}$ mice do not extinguish their fear. As this is one of the hallmarks of PTSD, a study in PTSD patients screened the genetic differences in the structure of the MR would be very interesting. In support, loss of function single nucleotide polymorphisms (SNP's) of the MR have shown enhanced susceptibility to a psychosocial challenge [64]. Studies on the influence of MR and GR on (catecholaminergic) stress responses and stress related pathologies have shown an correlation between genetic variance of MR and GR and the increased occurrence of PTSD and depression [64-67].

I expect that on the long term, the proposed lines of research will provide more insight in the development and treatment of PTSD. In parallel, we will gain more knowledge on how the glucocorticoid stress system affects the integration of emotion and cognition.

7.9 Conclusions
The following conclusions can be drawn:

1. Emotional contribution improves cognitive performance.

2. Both MR and GR activation influence the contribution of emotion to cognition.

3. Corticosterone treatment can have impairing and facilitating effects on emotional memory depending on the genetic background of the mice and the time of administration.

4. BALB/c and C57BL/6J mice are good models to study the role of the glucocorticoid stress system on stress related disorders such as PTSD.

5. The MR is a promising drug target that can be used for treating PTSD related pathology.

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