

# 5

## Propranolol reduces emotional distraction in working memory: A partial mediating role of propranolol-induced cortisol increases?

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## Abstract

Noradrenalin modulates prefrontal function, such as working memory (WM), and is associated with enhanced distractibility, and enhanced memory for emotional events and stimuli. The beta-blocker propranolol has been shown to reduce memory for emotional stimuli. Herein we describe investigations aimed at assessing whether the administration of propranolol would reduce the interference by emotional distractions during WM performance. In a between-subjects design, 48 young, healthy men received 80 mg propranolol ( $n = 25$ ) or placebo ( $n = 23$ ), before performing an “emotional Sternberg task” with neutral and negatively arousing distracters. Compared to placebo, propranolol impaired WM at low load, however, it also reduced the interference by emotional distracters at high load. Furthermore, an explorative moderated-mediation analysis indicated that the observed propranolol effects on emotional distraction were partially mediated by cortisol. In future non-clinical and clinical memory studies using propranolol administration, cortisol elevations should be monitored to further investigate the potential mediating role of cortisol.

## Introduction

When stressed, one of the neurohormonal systems that is activated is the locus coeruleus- noradrenergic system (Berridge & Waterhouse, 2003). This system plays a key modulatory role in prefrontal function (Berridge & Waterhouse, 2003; Minzenberg, Watrous, Yoon, Ursu, & Carter, 2008; Ramos & Arnsten, 2007), and is critically involved in emotional memory (McGaugh & Roozendaal, 2002; Roozendaal, Barsegyan, & Lee, 2008). Optimal levels of noradrenalin (NA) can improve functioning of the prefrontal cortex (PFC), whereas excessive NA or a depletion of NA impairs PFC function (Arnsten, 2009; Ramos & Arnsten, 2007). Stress-induced elevated NA is thought to take the reflective PFC “of-line” in favor of other more posterior brain areas, such as amygdala, hippocampus, and sensory- and motor areas, to allow for rapid emotional, or more habitual and reflexive behaviors (Arnsten, 1997; Arnsten, 1997; Ramos & Arnsten, 2007). Given the importance of the PFC in working memory (WM) performance (Kane & Engle, 2002; Ranganath et al., 2003), it is of no surprise that high levels of NA have also been found to be associated with impaired WM performance (Arnsten, Mathew, Ubriani, Taylor, & Li, 1999; Birnbaum, Gobeske, Auerbach, Taylor, & Arnsten, 1999; Mao, Arnsten, & Li, 1999).

WM can be defined as the capacity to maintain relevant information and to suppress irrelevant information. Patients with stress-related psychiatric disorders such as PTSD and depression, show poor WM performance and stronger interference from irrelevant negative emotional material (Joormann & Gotlib, 2008; Morey et al., 2009). Typically, in PTSD patients, pharmacological challenge tests or exposure to traumatic reminders are associated with increased noradrenergic responsiveness (Bremner, Krystal, Southwick, & Charney, 1996), and hypoactive responding in medial PFC, along with a hyperactive amygdala (Elzinga & Bremner, 2002; Etkin & Wager, 2007; Liberzon & Sripada, 2008; Shin, Rauch, & Pitman, 2006). When instructed to ignore emotional images shown during a WM task, PTSD patients displayed a similar pattern of decreased activity in dorsal areas, associated with WM and attention, and an enhanced neural activity in ventral areas (including the amygdala) associated with emotion processing relative to the trauma-exposed non-PTSD control group (Morey et al., 2009). These observations may be described as an exaggerated form of a “normal” response to emotional distractions during WM. That is, healthy

individuals also pay more attention to emotional stimuli than neutral ones, because of their salience and significance for survival even when these are deemed irrelevant, for example, in a context of an ‘emotional WM task’, where emotional stimuli are used as distracters (Kensinger & Corkin, 2003). As a result, WM performance slows down during the emotional distraction trials (Dolcos & McCarthy, 2006; Kensinger & Corkin, 2003).

The response to emotional stimuli by the amygdala is mediated by NA (Berridge & Waterhouse, 2003; van Stegeren et al., 2005; van Stegeren, Wolf, Everaerd, & Rombouts, 2008). Elevated NA enhances amygdala response (Onur et al., 2009) and enhances the attention for emotional stimuli (DeMartino, Strange, & Dolan, 2008). Imaging studies have shown that administration of propranolol, a highly lipophilic non-selective beta-adrenergic receptor blocker, that blocks the action of adrenalin on both beta1 and beta2 adrenergic receptors, reduces the activity in the amygdala during emotional processing (Strange & Dolan, 2004; van Stegeren et al., 2005). A number of studies aimed at elucidating the role of NA in emotional memory, have further shown that propranolol generally reduces memory for emotional events and stimuli (see for a review Chamberlain et al., 2006), when encoding takes place after propranolol administration (Cahill et al., 1994; Cahill & van Stegeren, 2003; van Stegeren et al., 2005). Taken together, these findings suggest that propranolol might improve emotional WM performance, owing to the diminished interference of emotional distractions.

The main aim of the present study was to investigate whether propranolol would improve emotional WM performance in young healthy men, by reducing the impact of emotionally negative distracters. Furthermore, we also performed an explorative analysis to investigate whether the stress hormone cortisol might mediate the effects of propranolol on emotional WM performance. There were two indicators that point towards a possible mediating role of cortisol in this regard: First, propranolol administration had been previously shown to elevate the levels of cortisol in the present sample (Tollenaar et al., 2009), as well as in other memory studies in which propranolol was administered (Maheu, Joober, Beaulieu, & Lupien, 2004; Maheu, Joober, & Lupien, 2005b). Secondly, as part of the present study, we have also found that cortisol administration leads to enhanced performance on the present emotional WM memory task (Oei, Tollenaar, Spinhoven, & Elzinga, 2009).

## Methods

### *Participants*

Male volunteers were recruited by means of a sign-up board and advertisements posted at the Faculty of Social and Behavioral Sciences of Leiden University. 54 participants who were part of a larger study on the effects of hydrocortisone and propranolol on memory functioning (Tollenaar et al., 2009) were included and randomly assigned to a propranolol and a control group in a double blind placebo-controlled between-subjects design (see Oei et al. 2009, for the study which compared hydrocortisone versus placebo). All participants had been screened before inclusion. Eligibility criteria were: no hypotension (blood pressure lower than 100/70 mmHg), no history of disease, no current use of prescribed medication or the use of remedies containing corticosteroids, no use of psychotropic drugs, no current and past psychiatric problems, a Body Mass Index (BMI; kg/m<sup>2</sup>) between 19 and 26, and age between 18 and 35 yrs. Each participant gave signed informed consent in which confidentiality, anonymity, and the opportunity to withdraw without penalty were assured. The experimental group received a fixed dose of 80 mg of propranolol (Inderal; peak 1-4 hr, halftime 3-6 hr), and the control group received placebo. Characteristics of the sample ( $n = 54$ ) were as follows (mean  $\pm$  SD): Age,  $20.13 \pm 1.92$  yrs; BMI,  $21.99 \pm 2.32$ ; trait anxiety as assessed with the State-Trait Anxiety Inventory (STAI, Spielberger, 1983),  $32.65 \pm 8.06$ ; WM estimate as measured with the subtest Digit Span Total score of the Wechsler Adult Intelligence Scale (WAIS) ,  $10.59 \pm 2.47$ ; psychoneuroticism, as assessed with the Symptom Checklist-90 (SCL-90, Arrindell & Ettema, 1986),  $118 \pm 22.08$ . The experimental group (mean age  $\pm$  SD resp.,  $20.74 \pm 2.21$ , range: 18 – 25 yrs) was older than the control group ( $19.52 \pm 1.37$  yrs, range: 18-24 yrs) ( $F[1, 53] = 5.96$ ,  $p = .02$ ). There were no other significant group differences (all  $ps > .05$ ). The Medical Ethical Committee of the Leiden University Medical Center approved of the study protocol, and it was carried out according to the standards of the Declaration of Helsinki (2000). Participants received course credit or a monetary compensation for taking part in the study.

### *Physiological recordings*

Cardiovascular measures. Systolic blood pressure (SBP, mmHg), diastolic blood pressure (DBP, mmHg), and heart rate (HR, bpm) were recorded using an automatic wrist blood pressure monitor (OMRON, R5-I).

Saliva sampling. Cortisol and  $\alpha$ -amylase were assessed via saliva samples (before pill ingestion, and before and after task performance at about peak propranolol levels) using Salivettes (Sarstedt, Germany). Saliva sampling is a stress-free method to assess unbound cortisol and  $\alpha$ -amylase (Kirschbaum & Hellhammer, 1994). Saliva samples were centrifuged and stored at  $-20\text{ }^{\circ}\text{C}$  until assayed at Prof Kirschbaum's laboratory (<http://biopsychologie.tu-dresden.de>). Cortisol and alpha-amylase concentrations in saliva were measured using a commercially available chemiluminescence-immunoassay kit with high sensitivity of 0.16 ng/ml (IBL, Hamburg, Germany). Inter- and intra-assay coefficients of variation were below 10 %.

### **Working memory task**

WM was measured using an adapted version of the Sternberg item-recognition task (Sternberg, 1966) previously described by Oei and colleagues (Oei et al., 2009). The WM processing load was manipulated by varying the numbers of uppercase letters (1 to 4 targets) that had to be held in memory for later recognition, and by varying the number of letters (1 to 4 displayed) presented in the recognition display after a short delay (1500-ms), which led to a load of 2 to 16 comparisons. For example, if the participant had to hold four items in memory (e.g., *E*, *R*, *F* and *S*), while searching for one of the items in a recognition display containing four items (*D*, *M*, *U*, and *Z*), this led to 16 possible comparisons (*E-D*, *E-M*, *E-U*, *E-Z*, *R-D*, *R-M*, *R-U*, *R-Z*, *S-D*, *S-M*, *S-U*, *F-D*, *F-M*, *F-U*, *F-Z* and *S-Z*). There were 3 blocks with low comparison load (load 2, 4, 6) and three with high comparison loads (load 8, 12, 16). In the delay-phase between target- and recognition display that originally contained a fixation cross (Lupien, et al., 1999; Oei, et al., 2006), distracters were presented that consisted of pictures selected from the International Affective Pictures System (Lang et al., 2001). Half of the distracters was emotionally neutral-, the other half was of negatively arousing content. A red fixation cross was shown at the centre of each picture. Participants had to ignore the distracters and press a 'yes' button indicating they had recognized a target (present-target trials), or a 'no' button, when no target letter was recognized (absent-target trials). Only one target letter was present in the present-target trials. Blocks with differing loads were randomly delivered. A total of 136 trials were delivered, which lasted approximately 10 minutes. Stimulus software (WESP) developed at the University of Amsterdam was used which randomizes and presents stimuli, and records reaction times and errors.

### **Procedure**

Participants were seated on a chair in front of a 17" CRT monitor with a fixed button box on the table before them. The first saliva sample was taken just before pill-ingestion. After pill ingestion, 75 minutes was spent reading magazines and filling out questionnaires. Then, cognitive tests were done for the larger study (for the entire procedure, see Tollenaar et al., 2009). At 110 min after the first saliva sample, a second sample was taken. Immediately hereafter, WM task instructions appeared on the computer screen. The task was first explained and participants were given the opportunity to practice the WM task in a short practice block, which consisted of 10 trials with only neutral distracters. Furthermore, they were asked to respond as quickly and accurately as possible. The last saliva sample was taken at 130 min after the first sampling, just after the WM task. At the end of the experiment, an exit interview was done in which it was asked whether participants thought they had been given placebo, or one of the study medications.

### **Statistics**

Reaction times were checked for errors, misses and outliers. Errors and misses were scored and removed. Outliers were replaced by the mean per load by arousal type + 2 standard deviations. Data (RTs and errors) of present and absent-target trials were separately analyzed using repeated measures ANOVAs, with as between-subjects factor Group (propranolol vs. placebo), Load (high vs. low) and Arousal (emotional vs. neutral) as within-subjects factors. Errors were analyzed likewise. Follow-up analysis of repeated measures ANOVA effects, if relevant, was done with *t*-tests. Physiological data were log-transformed when the assumption of normality was violated. Unpaired *t*-tests were done to test whether groups differed at the three time points. The data were analysed using SPSS for Windows, version 16.

## **Results**

WM data of two participants from the placebo group and one from the propranolol group were not recorded because of a computer failure. Three participants (two from the placebo group and one from the propranolol group) had to be excluded from further analyses because of extreme numbers of errors (>25%), leading to missing data in at least one category. A total of 48

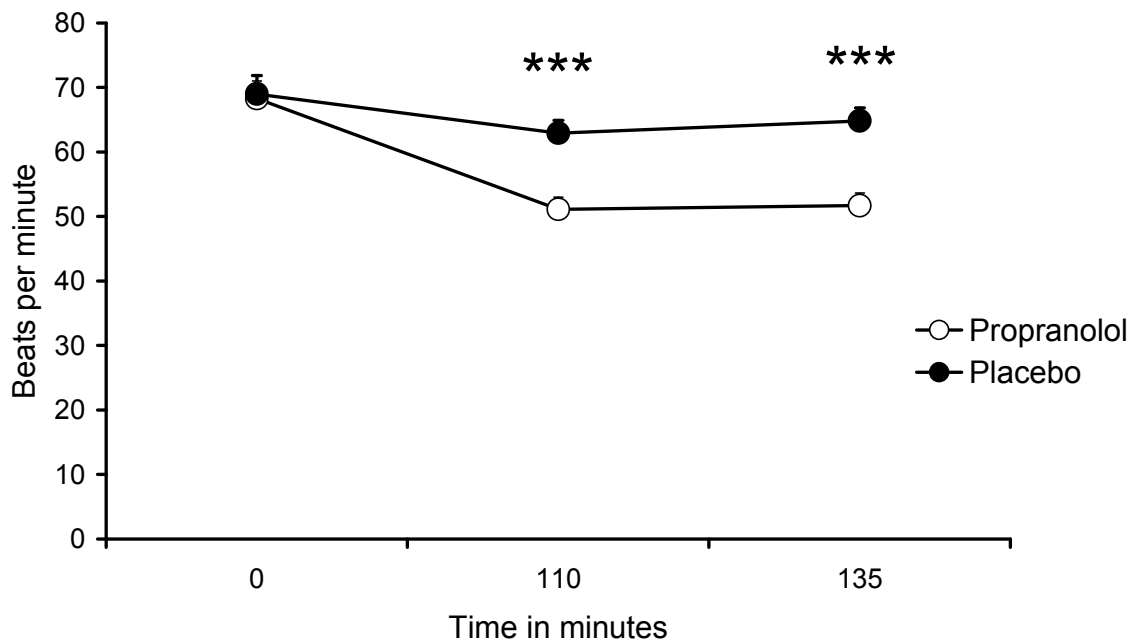
participants, 25 participants in the propranolol group and 23 in the placebo group were left for further analysis. Participants were not able to tell whether they had received placebo or propranolol (Chi-square = 4.70,  $df = 4$ ,  $p = .32$ ): Four participants correctly indicated noticing an effect of propranolol, while five participant, who received placebo, erroneously indicated that they had ingested propranolol

### Physiological measurements

Heart rate. See Figure 1 for means and standard errors. Separate  $t$ -tests showed that heart rate was significantly lower in the propranolol group compared to placebo, at t110 ( $t_{46} = 4.31$ ,  $p < .0005$ ) and t135 ( $t_{46} = 4.71$ ,  $p < .0005$ ), but not at baseline, t1 ( $t_{46} = 0.18$ ,  $p = .86$ ).

Blood pressure. Systolic blood pressure was significantly lower in the propranolol group after pill administration (systolic blood pressure at t1,  $t_{46} = 0.41$ ,  $p = .68$ ; t110,  $t_{46} = 2.83$ ,  $p = .007$ ; t135,  $t_{46} = 3.99$ ,  $p < .0005$ ; diastolic blood pressure showed a trend at t135,  $t_{46} = 1.91$ ,  $p = .06$ . At both other time points,  $ps > .36$ , see Figure 2 for means and standard errors).

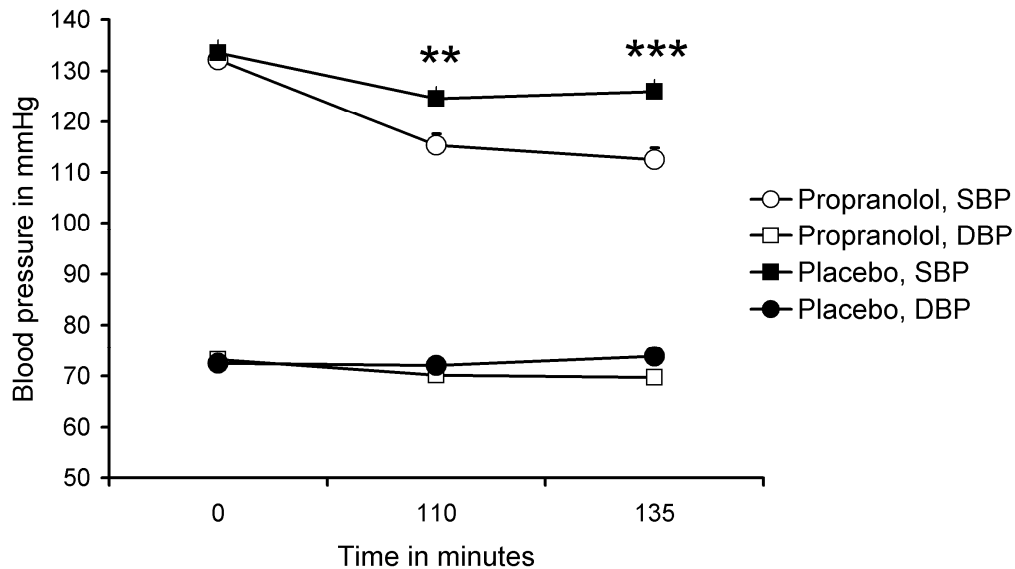
Figure 1. Mean heart rate and standard errors in the propranolol- and placebo group



\*\*\* = significant difference between groups,  $p < .0005$



**Figure 2.** Mean blood pressure and standard errors in the propranolol- and placebo group



*Note.* SBP = systolic blood pressure; DBP = diastolic blood pressure; significant difference between groups: \*\* =  $p < .01$ , \*\*\* =  $p < .0005$

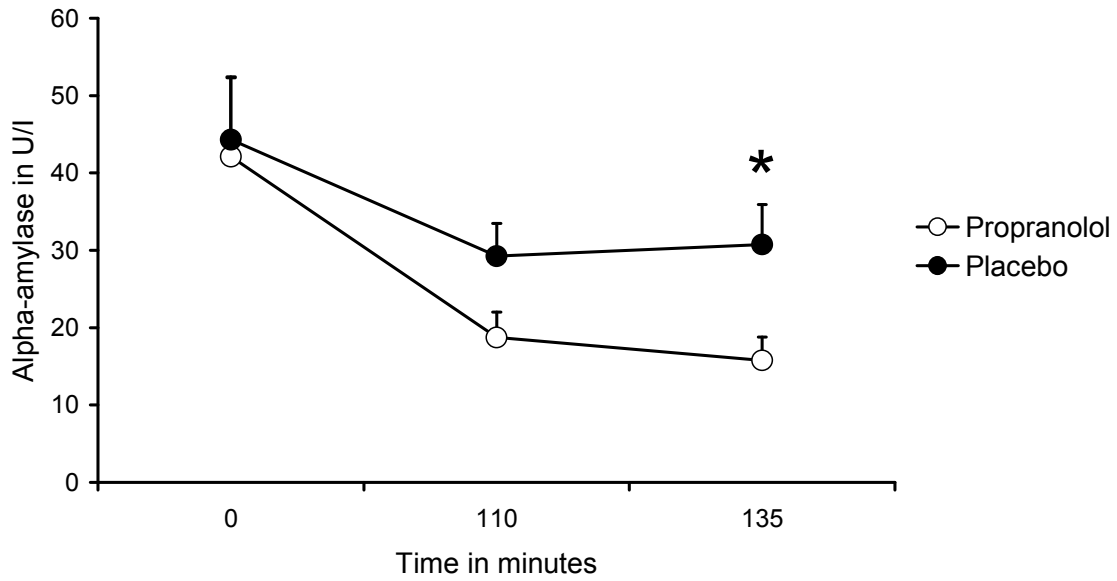
### Alpha-amylase

Amylase data were normalized using log-transformations (see Figure 3 for untransformed means and standard errors). There were no differences between groups at baseline just before pill administration ( $t_0$ ,  $t_{45} = 0.85$ ,  $p = .40$ ). The propranolol group had trend-level lower amylase levels just before WM testing ( $t_{110}$ ,  $t_{45} = 1.75$ ,  $p = .08$ ) and significantly lower values after testing ( $t_{135}$ ,  $t_{46} = 2.29$ ,  $p = .03$ ) as compared with the placebo group.

### Cortisol

Mean cortisol levels and standard errors at the three time points are depicted in Figure 4. At  $t_1$ , logtransformed cortisol levels did not differ between groups ( $t_{30.66} = 0.27$ ,  $p = .79$ ). The placebo group had lower cortisol levels at  $t_{110}$  ( $t_{46} = -3.29$ ,  $p = .002$ ), and  $t_{135}$  ( $t_{46} = -3.45$ ,  $p = .001$ ).

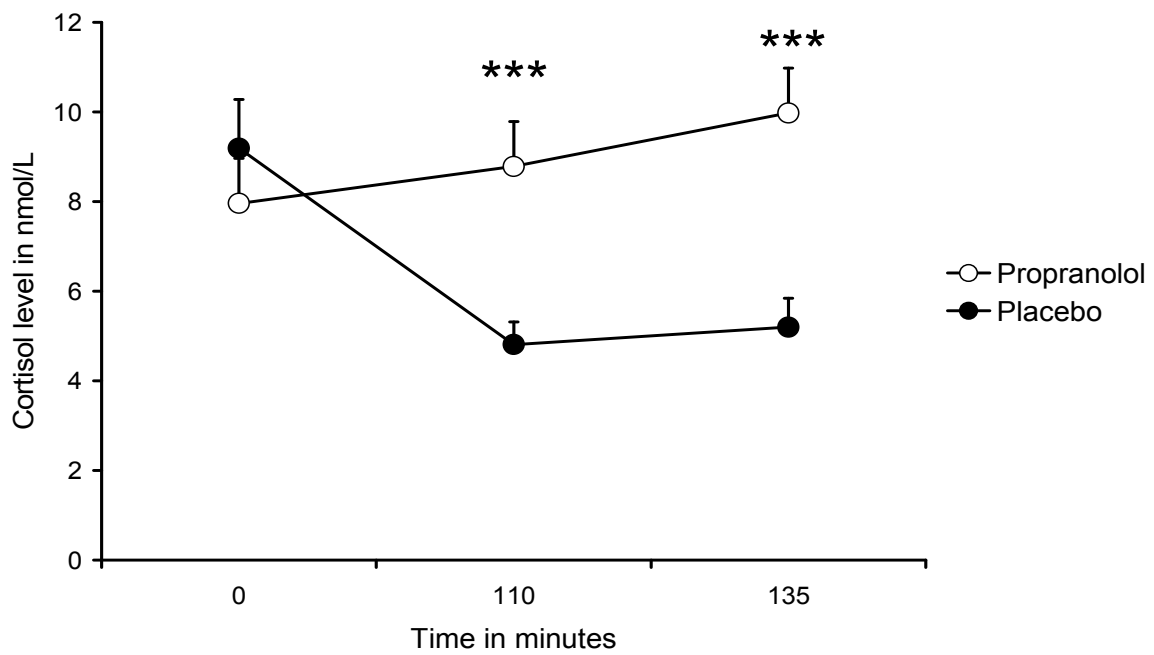
Figure 3. Mean alpha-amylase and standard errors in the propranolol- and placebo group



\* = significant difference between groups,  $p < .05$

Figure 4. Mean cortisol levels and standard errors in the propranolol- and placebo group

\*\*\* = significant difference between groups at  $p < .005$



## ***Emotional working memory performance***

### ***Present-target trials***

#### ***Reaction times.***

Mean reaction times and standard errors are shown in Table 1. At Present-target trials, significant within-subjects effects for Load ( $F[1, 46] = 293.51$ ) and Distracter ( $F[1, 46] = 25.79$ ) were found, with shorter RTs at low load compared to high load, and shorter RTs when distracters were neutral compared to emotional (both  $ps < .0005$ ). Also, a significant Group by Load by Distracter effect was revealed ( $F[1, 46] = 5.49, p = .02$ ), which indicated that in contrast to the propranolol group, the placebo group was significantly slower during emotional trials than neutral trials at high load (see Figure 5). There was, however, no significant overall between-groups effect ( $F[1, 46] = 0.62, p = .43$ ).

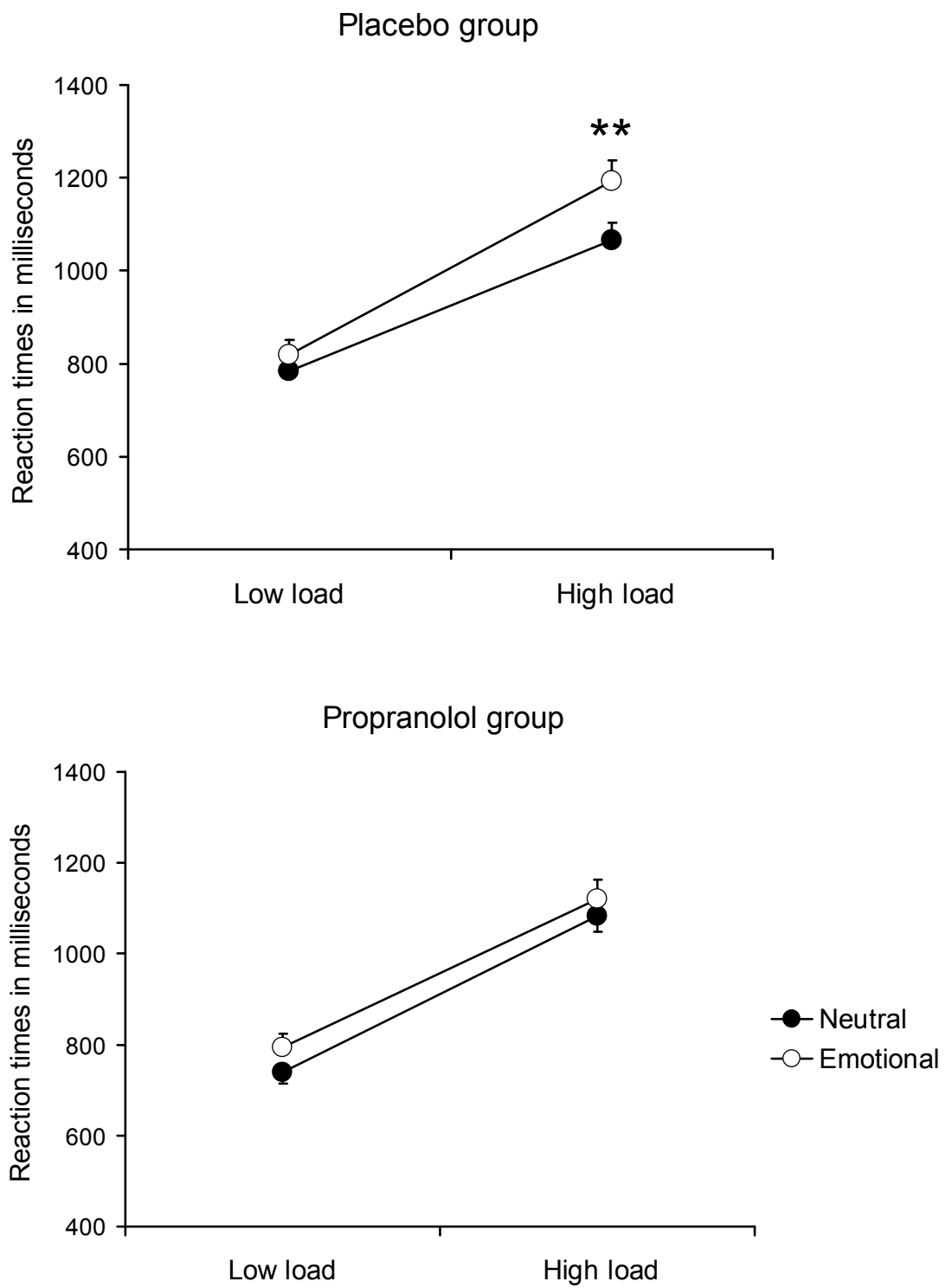
#### ***Errors.***

Mean errors (and *SE*) are shown in Table 2. Analysis of errors indicated that the significant triple interaction in RTs during present-target trials was not the result of a speed/accuracy trade-off (Group x Load x Distracter,  $F[1, 46] = 0.44, p = .51$ ). During present-target trials significantly less errors were made at low Load ( $M \pm SE, 0.73 \pm 0.08$ ), than at High Load ( $2.49 \pm 0.19$ ). There was an interaction between Group and Load ( $F[1, 46] = 5.19, p = .03$ ), which revealed that the propranolol group made more errors at low load ( $0.90 \pm 0.71$ ) than the placebo group ( $0.57 \pm 0.41$ ) ( $t_{38.92} = -2.03, p = .05$ ) but not at high load (placebo group:  $2.78 \pm 0.35$ ; propranolol group:  $2.20 \pm 0.18, t_{32.61} = 1.47, p = .15$ ). Furthermore, there was a within-subjects interaction between Load and Distracter  $F(1, 46) = 4.28, p = .02$ . At high load, more errors were made when distracters were emotional than when they were neutral. There were no other significant effects (all  $ps > .18$ ).

**Table 1. Means (*M*) and standard errors (*SE*) of the reaction times in the propranolol- and placebo group**

Group		Propranolol		Placebo	
Load		Low	High	Low	High
Target	Distracter	<i>M</i> ± <i>SE</i>	<i>M</i> ± <i>SE</i>	<i>M</i> ± <i>SE</i>	<i>M</i> ± <i>SE</i>
Present	Emotional	793.87 ± 29.84	1120.18 ± 41.34	819.94 ± 31.11	1192.96 ± 43.09
	Neutral	739.16 ± 26.28	1082.95 ± 34.27	785.13 ± 27.40	1067.03 ± 35.75
	Total	766.52 ± 26.85	1101.57 ± 35.14	802.54 ± 27.99	1129.99 ± 36.63
Absent	Emotional	848.79 ± 28.77	1261.38 ± 47.08	849.68 ± 29.99	1356.52 ± 49.08
	Neutral	808.03 ± 29.87	1196.74 ± 49.41	824.97 ± 31.15	1337.57 ± 51.52
	Total	828.41 ± 27.88	1229.06 ± 44.89	837.32 ± 29.06	1347.04 ± 46.81

Figure 5. The Group by Load by Distracter interaction



\*\*=difference between emotional and neutral trials within placebo group,  $t(22) = 3.55, p < .005$

**Table 2. Means and standard errors of the Error rates in the propranolol- and placebo group**

Group		Propranolol		Placebo	
Load		Low	High	Low	High
Target	Distracter	$M \pm SE$	$M \pm SE$	$M \pm SE$	$M \pm SE$
Present	Emotional	0.80 ± 0.16	2.48 ± 0.32	0.57 ± 0.16	3.00 ± 0.33
	Neutral	1.00 ± 0.10	1.92 ± 0.30	0.57 ± 0.17	2.57 ± 0.31
	Total	0.90 ± 0.12	2.20 ± .27	0.57 ± 0.12	2.78 ± 0.28
Absent	Emotional	0.56 ± 0.14	0.68 ± 0.19	0.44 ± 0.15	0.48 ± 0.19
	Neutral	0.52 ± 0.13	0.48 ± 0.18	0.44 ± 0.14	0.70 ± 0.18
	Total	0.54 ± 0.11	0.58 ± 0.14	0.44 ± 0.11	0.59 ± 0.15

### **Absent-target trials**

#### **Reaction times.**

See Table 1 for means and standard errors during absent-target trials. During absent-target trials, the within-subjects factors Load ( $F[1, 46] = 285.02, p < .0005$ ) and Distracter ( $F[1, 46] = 6.46, p = .01$ ) were significantly different, with faster RTs at low load ( $M \pm SE$ : Low load,  $832.87 \pm 20.14$ ; High load,  $1288.05 \pm 32.43$ ), and faster RTs when distracters were neutral ( $M \pm SE$ : emotional trials,  $1079.09 \pm 23.89$ ; neutral trials,  $1041 \pm 25.10$ ). There was a significant interaction between Group and Load ( $F[2, 46] = 4.09, p = .05$ ), with a trend for faster RTs in the propranolol group at high load ( $M \pm SE$ :  $1229.06 \pm 44.89$ ) compared to placebo ( $M \pm SE$ :  $1347.04 \pm 46.81$ ) ( $t_{46} = 1.82, p = .08$ ), but not at low load ( $t_{46} = 0.22, p = .83$ ). There was no significant overall difference between groups ( $F[1, 46] = 1.84, p = .18$ ), nor other interactions (all  $ps > .30$ ).

#### **Errors.**

Analysis of the errors during absent-target trials, revealed no significant within- or between-subjects effect, nor any interaction effects (all  $Fs < 1.59$ , all  $ps > .21$ ) (see Table 2 for means and standard errors).

### ***Explorative moderated-mediation analysis***

There were differences in cortisol levels between the propranolol and placebo group, with significantly decreasing cortisol levels over time in the placebo group only (see Tollenaar et al., 2009). Inspection of the data revealed that in the placebo group all - but one participant - showed decreased cortisol levels compared to baseline. In contrast, in the propranolol group, apart from 3 participants, there was an absence of decrease and in half of the group ( $n = 12$ ) cortisol levels even increased between baseline and the start of the WM task (maximum increase ( $t_{110} - t_0$ ) = 16.59 nmol/L).

To explore whether the enhancing effects of propranolol on the emotional trials of the WM task were mediated by cortisol, we first converted the RTs of the present-target trials into a single difference score (WMDiff), by subtracting the difference in RTs between High load emotional trials and neutral trials, from the difference between Low load emotional and neutral trials. This way, high scores represented a load-dependent difference between emotional and neutral trials, while low scores represented smaller differences in load and distracter type ( $M \pm SE$ : propranolol group,  $-12.13 \pm 25.49$ , placebo group,  $85.95 \pm 35.99$ ,  $F(1, 47) = 5.07$ ,  $p = .03$ ). Furthermore, a cortisol difference score was calculated (cortisol level just before testing WM minus baseline cortisol level, just before ingesting propranolol or placebo) ( $M \pm SE$ : propranolol group,  $0.82 \pm 1.18$ , placebo group,  $-4.38 \pm 0.96$ ,  $F(1, 47) = 11.42$ ,  $p = .001$ ). These two new variables were then checked for outliers. In both groups two outliers were detected which were subsequently removed (all outliers had extreme values regarding cortisol difference scores: placebo group outliers were due to extremes in baseline cortisol levels  $> 18$  nmol/L, the propranolol group outliers were the two participants with the highest increase in cortisol level after propranolol administration: increase  $> 12$  nmol/L, leading to cortisol levels  $> 16$  nmol/L).

As a next step, the dependent variable was entered into a moderated mediation model (see Preacher, Rucker, & Hayes, 2007) with Group as independent variable, and cortisol-difference score level as mediator variable, and Group as moderator variable. Group was added as moderator variable to be able to differentiate the conditional indirect effect of propranolol on the dependent variable, because the influence of placebo on cortisol is likely to be zero (Model 1, Preacher et al., 2007). The SPSS macro used, was provided by Dr. A. Hayes (<http://www.comm.ohio state.edu/ahayes/SPSS%20programs/modmed.htm>). It calculates the Sobel test for the conditional indirect effects as well as its percentile-based, bias-corrected, and bias-corrected and accelerated bootstrap confidence intervals, which is recommended for small samples (Preacher &

Hayes, 2004; Shrout & Bolger, 2002). Estimates of all paths were calculated using ordinary least squares regression. The results of these analyses are displayed in Table 3.

**Table 3. Regression results of the moderated mediation model**

Mediator Variable Model				
Predictor	<i>B</i>	<i>SE</i>	<i>t</i>	<i>p</i>
Constant	-3.53	0.89	-3.94	.0003
Group	3.14	1.23	2.54	.015
Dependent Variable Model				
Predictor	<i>B</i>	<i>SE</i>	<i>t</i>	<i>p</i>
Constant	113.44	42.81	2.65	.01
Cortisol	12.97	8.35	1.55	.13
Group	-129.31	52.19	-2.28	.02
Cortisol x Group	-26.43	10.19	-2.42	.02

A statistically significant interaction was found between Cortisol and Group, indicating that the indirect effect of Cortisol on WMdiff was moderated by group, with smaller WMdiff scores as cortisol levels increased. None of the bootstrap confidence intervals when Group was propranolol contained 0, which confirmed that the conditional indirect effect of propranolol was significant at  $\alpha = .05$  (see Table 4).



**Table 4. Bootstrapped conditional indirect effects of Group on WM score via Cortisol at specific values of the moderator (Group)**

Group	Point estimate	SE	Z	Bootstrapping					
				Percentile 95 CI		BC 95% CI		BCA 95% CI	
				Lower	Upper	Lower	Upper	Lower	Upper
Placebo	40.88	36.47	1.12	-20.40	126.01	-9.91	143.55	-11.75	140.74
Propranolol	-41.59	24.29	-1.71	-97.66	-2.43	-106.66	-6.85	-107.05	-6.97

**Note.** CI = confidence interval; BC = Bias-corrected; BCA = Bias-corrected and accelerated; 5.000 bootstrap samples

Finally, we investigated the possibility that propranolol-induced cortisol elevations were markers for inter-individual propranolol efficacy. An increased propranolol action along with cortisol elevations could mean that propranolol reduced emotional distracter interference due to a stronger reduction in adrenergic activation, rather than through cortisol release. However, post-hoc ANOVAs with the propranolol group divided into a high cortisol elevation ( $n = 12$ ) and a no elevation group ( $n = 13$ ), did not show any differences in change in levels of alpha-amylase, heart rate, or blood pressure, nor any significant correlation between cortisol and adrenergic measures (all  $ps > .05$ ).

## Discussion

In the present study the influence of 80 mg propranolol on performance of an adapted Sternberg WM task with emotional and neutral distracters at low and high comparison loads was investigated. Propranolol impaired WM with more errors at low load. However, consistent with our expectations, at high load, propranolol enhanced WM, with faster performance, indicating that propranolol reduced the distinction between emotional and neutral distracters at high load that was observed under placebo conditions. Furthermore, compared to placebo, administration of propranolol led to enhanced cortisol levels. Most interesting, cortisol appears to be involved in the decreased interference by emotional distraction following propranolol administration.

The finding that propranolol decreased the interference by emotional distraction in the present WM task, is in line with previous reviews describing the role of noradrenalin in arousal (Berridge et al., 2003, 2008) and the processing of arousing stimuli and amygdala activity (McGaugh & Roozendaal, 2002; Strange & Dolan, 2004; van Stegeren et al., 2005; van Stegeren, 2008). When administered before memory encoding, propranolol abolishes the arousal-induced enhanced memory for emotional events or stimuli, i.e., the “emotional memory effect” (van Stegeren, Everaerd, Cahill, McGaugh, & Gooren, 1998; Reist, Duffy, Fujimoto, & Cahill, 2001), however, without changing the subjective ratings of the emotional stimuli (Cahill et al., 1994). Similarly, in our study, propranolol may have weakened the arousal component of the emotionally negative distracters, thereby decreasing their interference, and consequently enhancing processing of these items. Another interpretation of the present results is that the attention for the emotionally negative distracters was

decreased. The detection of salient, motivationally relevant stimuli has been proposed to be modulated by the noradrenergic system (Strange & Dolan, 2007). In this regard, a recent functional imaging study has shown that activation in typical brain regions for (emotional and perceptual) oddball detection (ventrolateral PFC and temporoparietal junction) was abolished after propranolol administration (Strange & Dolan, 2007). There is, however, also evidence that the diminished attention after propranolol is regardless of item valence (DeMartino et al., 2008). Our results more likely suggest an effect specific to the emotional distracters, given the fact that we did not find overall diminished interference after propranolol.

Across several studies, propranolol was found to impair WM (Chamberlain et al., 2006). Here too, propranolol impaired WM performance at low load. Because NA has an inverted U shaped influence on WM, propranolol administration might specifically lead to impaired WM performance in individuals who have lower basal endogenous NA levels, as in low anxious individuals (Muller, Mottweiler, & Bublak, 2005). Apart from impairing effects, in contrast, beneficial effects of propranolol, were evident when comparing to performance in the placebo group under high cognitive load, when coping with emotional distraction. Consistent with the `load theory of selective attention` (Lavie et al., 2004; Lavie, 2005), our results showed that only at high cognitive load, the placebo group was slower when emotional distracters had to be ignored, compared to neutral distracters. According to this theory, there are enough cognitive control resources available to keep attention aimed at relevant stimuli when cognitive load is low, whereas a high load on cognitive control increases (irrelevant) distracter processing. The administration of propranolol, however, significantly reduced this effect. This might indicate that although propranolol appeared to impair prefrontal-dependent WM function, it might at the same time have attenuated amygdala-dependent processing of emotional stimuli, while functioning under heavy task demand.

As expected, propranolol inhibited the sympathetic nervous system (SNS), as assessed with adrenergic indices, such as heart rate, blood pressure, and alpha-amylase levels. Interestingly, the propranolol-treated group exhibited increased or unchanged cortisol levels rather than the typically observed decline of cortisol levels in the afternoon, as in the placebo group. Significant increases in cortisol levels following administration of 80 mg propranolol have been reported before (Kizildere, Gluck, Zietz, Scholmerich, & Straub, 2003; Lewis, Groom, Barber, & Henderson, 1981). In addition, significantly greater increases in cortisol levels have been observed in comparison to control conditions when propranolol

administration was followed by a stress-induction (Maheu et al., 2005b; Simeckova, Jansky, Lesna, Vybiral, & Sramek, 2000; but see Kudielka et al., 2007), physical exercise (Viru et al., 2007), a CRH challenge (Kizildere et al., 2003), or pentagastrin (Khan, Liberzon, & Abelson, 2004). These data all indicate that propranolol enhances HPA-axis activity, and suggest that the SNS via beta-adrenergic receptors, under such physically or socially stressful conditions –but without propranolol, decreases cortisol (Kizildere et al., 2003). The underlying mechanism responsible for these effects, however, remains unclear. Viru and colleagues (2007) proposed two possible mechanisms by which propranolol increases cortisol, (1) a dual excitatory and inhibitory role being played by the SNS on adrenocortical function, with inhibitory effects to avoid exaggerated hormonal responses, or (2) an adjustment which occurs solely in response to beta-blockade, through which increased adrenalin production compensates for decreased influence of noradrenalin, that enhances central CRH and ACTH levels, which in turn stimulate cortisol release. Further investigations into the observed increase in cortisol levels following propranolol administration are warranted and may serve to further clarify the mechanism(s) responsible for these effects.

Apart from the explanations for the direct effects of propranolol on emotional WM, we found evidence for indirect effects of propranolol by enhancing cortisol levels: Cortisol was at least partially involved in decreasing distracter interference of emotional stimuli. Propranolol-induced cortisol increases were related to less interference from emotional distractions. Similar effects of cortisol have been reported and are consistent with the present findings. Cortisol administration specifically reduced the distraction of emotional stimuli thereby enhancing performance in the same emotional WM task (Oei et al., 2009). Cortisol administration has also been shown to reduce selective attention to fear-related stimuli (Putman et al., 2007), to decrease startle reflex in response to valenced pictures (Buchanan et al., 2001), and to attenuate yohimbine-induced panic symptoms (Vasa et al., 2009).

At present, it is unclear how our finding that cortisol partially mediates emotional WM may be related to previous animal studies, which suggest that the (memory impairing) effects of administered glucocorticoids are abolished when adrenergic activation is blocked with beta-blockers (Okuda et al., 2004; Roozendaal et al., 2004a; Roozendaal et al., 2004b; Roozendaal et al., 2004c), or when lesions to the amygdala are inflicted (Roozendaal, Portillo-Marquez, & McGaugh, 1996; Roozendaal & McGaugh, 1997; Roozendaal et al., 2003). Two recent human studies are consistent with these animal studies, showing that

propranolol abolishes the effects of cortisone or stress on memory retrieval (de Quervain et al., 2007; Schwabe et al., 2009). Glucocorticoids effects on (human) WM might also depend on concurrent stress-induced (nor)adrenergic activity (Elzinga & Roelofs, 2005; Schoofs et al., 2008). However, in these latter two studies, adrenergic activity was not pharmacologically reduced, and only neutral material was used.

Ours and other reports of propranolol-induced cortisol elevations (Khan et al., 2004; Kizildere et al., 2003; Lewis et al., 1981; Maheu et al., 2004; Maheu et al., 2005b; Simeckova et al., 2000; Viru et al., 2007) suggest, however, that further investigations into propranolol-induced cortisol elevations are warranted in future memory studies. Propranolol-induced rises in cortisol levels, or inhibition of decreases in cortisol levels, as in the present study, are highly relevant to memory research in a broader sense, given the mounting body of evidence that supports a role for cortisol effects on memory (Buchanan & Lovallo, 2001; Buchanan et al., 2006; Cahill et al., 2003; de Quervain et al., 2000; Kuhlmann et al., 2005b; Oei et al., 2006; Tollenaar et al., 2009; Wolf et al., 2004).

The results of the present study are limited, because they represent the observed effects within a small group of healthy young men. Such a group is not necessarily indicative of the general population and our conclusions at this time are therefore somewhat confined.

Nevertheless, the present results indicate that propranolol administration reduced the interference by emotional distraction under high cognitive load. Our findings suggest that the reduction of irrelevant emotional interference might be one of the mechanisms underlying the therapeutic efficacy of propranolol in the treatment of stress-related psychiatric disorders (Brunet et al., 2008; Pitman et al., 2002; Pitman & Delahanty, 2005). Future studies aimed at the possible indirect effects elicited by propranolol via cortisol changes, may serve to clarify the mechanism(s) by which propranolol exerts its effects on memory.

