Available online at www.sciencedirect.com
ScienceDirect
Biological Psychology 73 (2006) 262–271
BIOLOGICAL PSYCHOLOGY
www.elsevier.com/locate/biopsycho

Effects of beta blockade, PTSD diagnosis, and explicit threat on the extinction and retention of an aversively conditioned response

Scott P. Orr [a,b,*], Mohammed R. Milad [b], Linda J. Metzger [a,b], Natasha B. Lasko [a,b], Mark W. Gilbertson [a], Roger K. Pitman [b]

[a] Department of Veterans Affairs Medical Center, Research Service (151), 718 Smyth Road, Manchester, NH 03104, United States
[b] Department of Psychiatry, Massachusetts General Hospital and Harvard Medical School, 149 13th Street, CNY 2618, Charlestown, MA 02129, United States

Received 13 June 2005; accepted 6 May 2006
Available online 7 July 2006

Abstract

An aversively conditioned SC response was assessed in 18 males meeting DSM-IV criteria for chronic posttraumatic stress disorder (PTSD) and 10 trauma-exposed males who never developed PTSD. Effects of beta blockade on acquisition and retention of a conditioned response (CR) were examined by administering propranolol HCl before acquisition or following extinction trials. Retention of the CR was assessed 1 week following acquisition under conditions of non-threat and threat. Conditioned stimuli were colored circles and the unconditioned stimulus (UCS) was a ''highly annoying'' electrical stimulus. The propranolol failed to produce any measurable effects on acquisition or retention of the CR and there was no evidence of increased conditionability in individuals diagnosed with PTSD. One week following acquisition, the differential CR to the reinforced stimulus was evident only in the threat condition. This suggests that belief in the presence of a threat is necessary and sufficient for activating a previously established CR.

Keywords: Conditioning; Extinction; Retention; Skin conductance; Posttraumatic stress disorder; Beta blockade

1. Introduction

It is well known that memories for stressful and emotional events become more strongly established than memories for neutral events (McGaugh and Roozendaal, 2002). Intense emotional responses that accompany a traumatic event may help to explain why individuals with posttraumatic stress disorder (PTSD) develop strong memories for the experience. Memories for traumatic events can persist for many years, even decades (e.g., Orr et al., 1993). This ''over-consolidation'' may produce emotional memories that are highly resistant to extinction (Orr et al., 2000; Pitman et al., 2001). The ability to manipulate the stress hormones that are important to memory consolidation, perhaps immediately after a traumatic event, could reduce an individual's risk of developing anxiety disorders, such as PTSD.

The facilitation of emotional memory consolidation has been demonstrated to be influenced by adrenal hormones, i.e., catecholamines and glucocorticoids. These hormones are secreted after a stressful experience and appear to influence noradrenergic activity of the basolateral amygdala; the memory enhancing effects of glucocorticoids appear to primarily involve the hippocampus, but require input from the basolateral amygdala (see Roozendaal, 2002). Using an inhibitory avoidance procedure in animals, McGaugh and colleagues have shown that administration of propranolol, a b-adrenergic blocker, prior to, or immediately after, training inhibited the enhancement effects of norepinephrine on memory consolidation (Cahill and McGaugh, 1996; Miranda et al., 2003; Salinas et al., 1997). Similarly, injections of norepinephrine prior to training facilitated memory consolidation (Hatfield and McGaugh, 1999; Introini-Collison et al., 1992; Sullivan et al., 1991). Blockade of b-receptors with propranolol in animals also abolished memory facilitation in spatial learning tasks (Hatfield and McGaugh, 1999; Ji et al., 2003b), eye-blink conditioning (Gould, 1998; Ji et al., 2003a), as well as contextual fear conditioning in animals (Roozendaal et al., 2004).

* Corresponding author. Tel.: +1 603 624 4366x6733; fax: +1 603 629 3265.
E-mail address: scott.orr@med.va.gov (S.P. Orr).

0301-0511/$ – see front matter
doi:10.1016/j.biopsycho.2006.05.001

Accumulating evidence from the human literature has begun to support the influence of norepinephrine on memory modulation. Cahill et al. (1994) showed that administration of propranolol blocked the memory enhancing effects of emotional arousal. Several other studies in humans have shown similar effects of propranolol on memory consolidation (e.g., Harmer et al., 2001). Propranolol has recently been found to influence conditioning to a fear context, but not to specific fear cues in healthy humans. In this study, Grillon et al. (2004) observed that individuals given propranolol prior to conditioning trials showed reduced SC levels when reexposed to the conditioning context. However, the magnitude of the SC conditioned response, during both acquisition and retention testing, was not influenced by propranolol.

A previous study by Orr et al. (2000) examined the acquisition and extinction of an aversively conditioned response in trauma-exposed individuals with and without PTSD. Results from this study demonstrated that individuals with, compared to without, PTSD produced larger SC responses to a stimulus paired with a mild shock (CS+) relative to a CS not paired with shock (CS−) during acquisition trials. The PTSD group's larger differential SC response to CS+ versus CS− trials persisted during extinction, even though participants had been told that they would no longer receive the electric shocks. Given that propranolol can reduce the memory enhancing effects of emotional arousal, it seems reasonable to expect that it might also reduce or eliminate the stronger conditioned responses (CRs) produced by some individuals. This prediction is based on the assumption that b-adrenergic activity plays a key role in the acquisition and extinction of conditioned emotional responses. However, the findings of Grillon et al. (2004) suggest that although b-adrenergic activity influences emotional memories, it may not influence the formation of conditioned emotional responses to specific fear cues.

The possibility that propranolol might retroactively interfere with the consolidation of a CR in PTSD is of particular interest. Might the administration of propranolol post-conditioning reduce the strength or durability of the CR? It is logical to hypothesize that if propranolol is present and exerting its actions from the start, as when it is administered prior to conditioning, it will be more effective. However, with regard to the clinical problem of PTSD, the potential advantages of an agent that could exert a beneficial effect when given following a traumatic event are obvious. Limited support for this possibility has been provided by findings from a study that examined the effect of administering propranolol shortly after a traumatic event on the subsequent development of PTSD (Pitman et al., 2002). Although propranolol did not prevent diagnosable PTSD from occurring, individuals who received propranolol showed significantly reduced physiological responses during mental imagery of the traumatic event, compared to those who did not receive it, when assessed several weeks after the event.

The study reported herein used a differential conditioning procedure to assess the acquisition and extinction of a skin conductance CR to emotionally neutral CSs (colored circles) paired with a mild electric shock UCS, as was done previously by Orr et al. (2000). The present study expands upon previous work in three ways. First, the study was designed to examine whether the administration of a single dose of propranolol before or after conditioning would influence the acquisition, extinction, and retention of an aversive CR, and whether it would diminish the heightened conditionability previously observed in PTSD. Second, durability of a CR established in the laboratory was examined by assessing the strength of the CR 1 week following conditioning. Third, the capacity of threat to modulate or even reinstate the experimental CR was tested by examining the CR under conditions where there clearly was or was not a possibility of receiving a shock UCS.

A potential problem with employing a b-adrenergic blocker to influence a central nervous system (CNS) process such as conditioning is that, because of its peripheral effects on the sympathetic nervous system (SNS), the presence of the drug could confound measurement of the peripheral-dependent variables that are being used to infer the central process the drug is putatively influencing. In this regard, measurement of skin conductance confers a distinct advantage. Although SC is unquestionably increased by central SNS activation, and in fact is probably one of the better dependent measures of this phenomenon, its peripheral action does not involve epinephrine or norepinephrine. Rather, both its preganglionic and postganglionic innervations are cholinergic. This fortunate consideration means that the conditioned SC response measured in the reported work may reasonably be expected to reflect propranolol's effect on central CR acquisition, extinction, and retention, free of peripheral contamination by the drug itself.

2. Methods

2.1. Participants

The sample consisted of 28 males with exposure to combat ($n = 25$) or firefighting ($n = 3$). Participants were recruited from the population of active and former VA outpatients, Vet Center clients, Professional Firefighters of New Hampshire, and posted notices at local fire stations. The research project was reviewed and approved by the Institutional Review Board of the Veterans Affairs Medical Center, Manchester, NH. Each research candidate underwent a diagnostic interview for the presence of Axis I mental disorders utilizing the structured clinical interview for DSM-IV (SCID; First et al., 1997). No candidate met DSM-IV criteria for organic mental disorder, schizophrenia, or current manic syndrome. On the basis of the Clinician-Administered PTSD scale (CAPS-I, Blake et al., 1995), participants who met DSM-IV criteria for current PTSD were classified into the PTSD group ($n = 18$); those who did not meet criteria for current or past PTSD were classified into the non-PTSD group ($n = 10$). Sixteen individuals in the PTSD group had one or more current comorbid Axis I disorders, as follows: 11 major depression, 1 bipolar, 3 dysthymia, 6 panic, 4 specific phobia, and 1 obsessive-compulsive. None of the non-PTSD participants had any current Axis I disorders. Fourteen of 18 (78%; Table 1) participants in the PTSD group, and 5 of 10 (50%) participants in the non-PTSD group, were determined to be using a psychoactive medication or drug at the time of psychophysiologic testing, as determined from self-report and/or urine drug screen. All participants were determined, on the basis of verbal report and results of an electrocardiogram, to be free from any medical condition that would preclude taking a single 40 mg oral dose of propranolol HCl.

Table 1
List of psychoactive medications (and daily dosages) as reported by participants in the PTSD group

Subject no.	Medication (daily dosage)
1	Sertraline (unknown), Rispiridone (unknown)
2	Trazodone (30 mg), Lithium (600 mg)
3	Bupropion (300 mg), Verapamil (240 mg), Temazepam (10 mg), Lotensin (10 mg)
4	Lorazepam (unknown), Trazodone (unknown)
5	None
6	None
7	None
8	Fluvoxamine (.50 mg), Zolpidem (10 mg), Buspirone (400 mg)
9	Buspirone (30 mg), Quetiapine (100 mg)
10	Trazodone (200 mg)
11	None
12	Prinivil (unknown), Lorazepam (unknown), Zolpidem (unknown), Paroxetine (unknown), Hydrochlorothiazide (unknown)
13	Bupropion (450 mg), Zolpidem (unknown)
14	Alprazolam (unknown), Atenolol (unknown), Sertraline (unknown), Trazodone (unknown), Isosorbide dinitrate (unknown)
15	Cyclobenzaprine (10 mg), Clonazepam (1 mg), Trazodone (150 mg)
16	Naltrexone (unknown)
17	None
18	Trazodone (50 mg)

2.2. Measures

2.2.1. Psychometric

These included the impact of event scale-revised (Weiss and Marmar, 1997), state-trait anxiety inventory (Spielberger et al., 1990), Beck depression inventory (BDI; Beck et al., 1979), and symptom checklist-90-revised (SCL-90-R; Derogatis, 1983).

2.2.2. Psychophysiologic

A Coulbourn Modular Instrument System was used to record SC level. Skin conductance was measured directly by a Coulbourn Isolated Skin Conductance coupler (S71-23) using a constant .5 V through 9 mm (sensor diameter) Sensor Medics Ag/AgCl electrodes placed on the hypothenar surface of the participant's non-dominant hand in accordance with published guidelines (Fowles et al., 1981). The SC electrodes were separated by 14 mm, as determined by the width of the adhesive collar. The SC level analog signal was digitized by a Coulbourn Lablinc Analog to Digital Converter (L25-12). An IBM-compatible computer system was utilized for sampling and storing the digitized signal.

2.3. Procedure

Details of the various study procedures were explained in full to participants during their initial study visit; their questions were answered and written informed consent obtained. Participants were interviewed regarding their health and any medical conditions that might contraindicate taking a single dose of propranolol. An electrocardiogram was performed in order to rule out the presence of cardiac abnormalities that might also contraindicate study participation. Once a participant was medically cleared, the structured clinical interview was performed and psychometric instruments administered. Following completion of the interview and psychometrics, participants determined the level of UCS to be used during the subsequent conditioning and retention sessions. The UCS was a 500 ms electrical pulse generated by a Coulbourn Transcutaneous Aversive Finger Stimulator (E13-22), which was isolated from line current and used a 9 V dry cell battery attached to an adjustable step-up transformer. After attaching electrodes to the second and third fingers of the dominant hand, the technician gave the following instructions:

> "For this experiment, you will set your own level of electric stimulation. You should choose a level that is highly annoying but not painful. I will start the stimulation at a very low level and gradually increase the level until you say 'stop.' The level that you set will then be used in a later part of the experiment."

The technician then proceeded to set the UCS level and noted the final dial setting of the transformer, which ranged between .2 and 4.0 mA. This value provided a measure of the UCS intensity for each individual.

Approximately 1 week following the initial visit the participant returned to the psychophysiology laboratory to undergo the conditioning procedure. Upon arrival at the laboratory, the participant was reminded that a mild electric stimulus would be used and that he was free to terminate the experiment at any time. One hour prior to entering the laboratory, the subject received a double-blind 40 mg oral dose of propranolol HCl or look-alike placebo prepared in advance and administered by either a physician or research nurse. Following the last extinction trial, the participant received a second double-blind 40 mg oral dose of propranolol HCl or look-alike placebo. However, no participant received two doses of propranolol. Thus, participants were randomly assigned to one of three drug conditions: placebo/placebo, in which propranolol was received neither before nor after conditioning; propranolol/placebo, in which propranolol was received before conditioning; or placebo/propranolol, in which propranolol was received after conditioning. Blood pressure and pulse were measured prior to the first dose of study medication, 1 h later (just prior to starting the conditioning procedure), and 1 h after receiving the second dose of study medication (following the last extinction trial).

The experimental session took place in a humidity- and temperature-controlled, sound-attenuated room, connected via wires to an adjoining laboratory in which the experimental apparatus was located. The participant was seated in a comfortable armchair and monitored via an unobtrusive video camera. The CS+ and CS− were represented by two differently colored 6 in. diameter circles, randomly selected for each participant from four options (red, blue, white, or green). The colored CSs were computer-generated and displayed on a monitor positioned 4 ft in front of the participant. A 500 ms electric shock previously determined by the subject to be "highly annoying but not painful" served as the UCS.

Physiologic recording electrodes and those for administering the electrical UCS were then attached. Next the participant was given the following instructions:

> "This experiment will consist of a baseline period followed by three phases. During the baseline period, which will last 5 min, we will check our instruments and you should try to relax. At the end of this period, you will see 'Begin Phase I' displayed on the monitor. During this phase, two different colored circles will be presented on the monitor. You should sit quietly and look at each colored circle as it is presented. At the end of the period, 'Begin Phase II' will appear on the monitor. During this phase, the colored circles will be presented again, and some of them will be followed by the electrical stimulus. Again, you should sit quietly and look at each colored circle as it is presented. At the end of Phase II, 'Begin Phase III' will appear on the monitor. During this phase, you will see more colored circles. However, you will no longer receive any electrical stimulation. Please continue to sit quietly and look at each colored circle as it is presented. It is important that you watch the screen at all times. Do you have any questions?"

When the participant was ready to proceed, the technician left the room and activated the computer, which took over the administration of the experiment. There was a 5 min baseline recording period during which the dependent measures were sampled at 2 Hz.

The Habituation phase (phase I) consisted of five presentations each of the to-be CS+ and CS− in pseudo-random order, i.e., there were no more than two consecutive presentations of the same stimulus type. The CS duration was 8 s, and the intertrial interval (ITI) was 20 ± 5 s, determined at random by the computer. During the acquisition phase (phase II) a 500 ms shock pulse occurred immediately following each CS+ offset. There were five presentations of each stimulus type. The extinction phase (phase III) consisted of 10 non-reinforced presentations

of the CS+ and CS−. The dependent physiologic measures were sampled at 10 Hz beginning 2 s prior to CS onset and ending 6 s following CS offset.

At the completion of the extinction phase, the technician re-entered the participant's room, removed the physiologic recording electrodes, and the second dose of study drug was administered. Participants were asked whether they were able to predict when the shock would occur and to identify the particular color of the circle that was paired with the shock, if this information was not spontaneously given.

Approximately 1 week later the participant returned to the laboratory to undergo the retention procedure. Retention of conditioned responses to the previously used CS+ and CS− was examined under two conditions: non-threat, followed by threat. Both conditions and the accompanying instructions were the same as for phase III (extinction) of the conditioning session, i.e., there were 10 presentations each of the previously used CS+ and CS− during each retention condition. During the non-threat retention condition, no shock electrodes were attached and the subject was informed that there would be no possibility of receiving any electrical stimuli. During the threat retention condition, shock electrodes were attached and the subject was informed that he might receive the electrical stimulus. The 500 ms shock was presented once, viz., following the fifth presentation of the CS+, during the threat retention condition.

Upon completion of the third session, participants were thanked and debriefed with regard to the general purpose of the experiment and any clinically relevant findings in their case.

2.3.1. Skin conductance scores

2.3.1.1. Response scores for CS and UCS intervals. A SC response score for each CS interval was calculated by subtracting the mean level for the 2 s immediately preceding CS onset from the highest value among those recorded during the 8 s CS interval. A SC response score for the interval containing the UCR was calculated by subtracting the average SC level within 6–8 s following CS onset, from the maximum increase in SC level during the .5–6.5 s interval following CS offset (which corresponded to the onset of the .5 s UCS). These scoring methods are the same as those published in our previous work (Orr et al., 2000).

2.3.1.2. Context conditioning score. In order to assess conditioning to the laboratory context, a measure of autonomic arousal was obtained from the mean SC level for the 2 s immediately preceding CS onset. These SC values were averaged for the first five CS+ and first five CS− trials for each of the three series of extinction trials, thereby creating separate values for each set of extinction trials. Anxiety or fear that is associated with a particular context would be expected to influence tonic SC level, which would be reflected in the pre-stimulus SC value.

3. Results

3.1. Demographic, psychometric, debriefing, UCS level and SC resting level, orienting response and unconditioned response

Means, S.D.s, and results of *t*-test comparisons between the PTSD and non-PTSD groups for the various demographic and psychometric measures, the UCS level set by the participant, SC resting level, orienting response and response to the UCS are presented in Table 2. As can be seen, the groups did not differ in age or education level. As expected, the PTSD group showed significantly more PTSD-specific and general psychopathology than the non-PTSD group. The PTSD and non-PTSD groups did not differ in their resting SC level during the initial 5 min resting baseline, and selected fairly comparable levels of the UCS. The mean SC response to the UCS for the PTSD group appears to be slightly larger than that for the non-PTSD group, although this difference did not approach statistical significance. During questioning, 20 of 27 individuals correctly identified the relationship between the CS+ and shock (contingency awareness was not assessed for one participant).

Table 2
PTSD and non-PTSD group means, standard deviations, and *t*-test results for the demographic and psychometric measures and resting physiological levels

	PTSD ($n = 18$)		Non-PTSD ($n = 10$)		*t*-Tests	
Measure	*M*	S.D.	*M*	S.D.	*t*(26)	*p*
Age	52.7	5.7	57.2	9.5	1.6	.13
Education	14.5	3.8	15.3	1.8	.7	.52
Impact of event scale						
Intrusion (0–35)	24.2	9.6	1.1	7.4	5.0	<.001
Avoidance (0–40)	22.5	8.9	4.0	6.6	5.7	<.001
Arousal (0–35)	24.7	10.3	5.7	7.6	5.1	<.001
CAPS total score (0–136)	71.4	20.9	5.3	7.9	9.6	<.001
STAI						
Trait (20–80)	54.4	12.2	21.8	10.1	5.4	<.001
State (20–80)	51.1	12.1	23.2	11.5	5.5	<.001
SCL-90-R, GSI (0–4)	1.6	.8	.4	.4	4.7	<.001
BDI (0–63)	25.2	11.9	7.2	6.3	4.4	<.001
Level of UCS (0–4)	2.6	.9	2.9	1.0	.7	.48
Skin conductance measures (μS)						
Resting level	5.4	3.7	6.0	10.0	.2	.81
OR	.4	.4	.2	.5	.7	.49
UCR	1.0	.6	.7	.5	1.7	.11

Note: PTSD = posttraumatic stress disorder; CAPS = clinician administered PTSD scale; STAI = state-trait anxiety inventory; SCL-90-R = symptom checklist-90-revised; GSI = global severity index; BDI = Beck depression inventory; UCS = unconditioned stimulus; resting level = mean during 5 min rest period; OR = orienting response, or averaged response to first presentation of the CS+ and CS− during the habituation phase; UCR = averaged unconditioned response for CS+ trials during the acquisition phase.

3.2. Effect of propranolol on HR level

The influence of propranolol on resting HR was examined by means of analysis of variance for repeated measures (ANOVAR) that included two factors: condition (propranolol, no propranolol), which was analyzed as a between-subjects effect, and time, which served as the repeated measure. The time factor contained three levels that corresponded to the resting HR level: (1) prior to drug ingestion, (2) 1 h following drug ingestion, and (3) following the last trial of the extinction phase. For the purpose of these analyses, HR data from the placebo/propranolol group were combined with those from the placebo/placebo group because the former group did not receive propranolol until after the third HR measurement. The resting HR levels (BPM) and S.D.s for the three time points in the no propranolol condition are: $M = 72.2$, S.D. = 12.0; $M = 63.5$, S.D. = 10.5; $M = 64.5$, S.D. = 9.5, respectively, and for the combined propranolol conditions are: $M = 72.9$, S.D. = 12.9; $M = 61.2$, S.D. = 10.2; $M = 58.3$, S.D. = 8.1, respectively. Results of ANOVAR yielded a non-significant condition main effect ($F(1, 24) < 1$) and condition × time interaction ($F(2, 48) = 2.4$, $p = .12$). The resting HR level declined over measurement periods as indicated by a significant time main effect ($F(2, 48) = 30.9$, $p < .001$).

In order to provide a more powerful examination of whether the dose of propranolol produced a discernable HR effect, the condition × time interaction was decomposed using the SAS profile option (SAS Institute, 1982), which allows for testing differences and interactions between adjacent means. This examination yielded a significant interaction effect between the second and third resting HR levels ($F(1, 24) = 6.2$, $p = .02$), indicating that there was a modest reduction in the later resting HR level in the group that received propranolol. There was no evidence of any condition × time interaction between the first and second HR measurements ($F(1, 24) < 1$).

3.3. Conditioning procedure

Analyses of variance for repeated measures were separately conducted for the habituation, acquisition, extinction, and retention phases. For each phase, two sets of analyses were initially performed in order to examine the effects of PTSD diagnosis and administration of propranolol. In the absence of any significant main effects or interactions associated with PTSD diagnosis and propranolol, a third set of analyses was performed that eliminated these effects from the ANOVAR model. Conditioning effects associated with the PTSD diagnosis were examined by an ANOVAR model that included three factors: diagnosis (PTSD, non-PTSD), which was analyzed as a between-subjects effect, stimulus type (CS+, CS−), which was analyzed as a within-subjects effect, and trials, which served as the repeated measure. The trials factor contained 5 levels (5 CS+ and 5 CS−) for habituation and acquisition phases, and 10 levels for the extinction and retention phases. Conditioning effects associated with propranolol administration were examined by an ANOVAR model that included: condition (placebo/placebo, propranolol/placebo, placebo/propranolol), which was analyzed as a between-subjects effect, and stimulus type (CS+, CS−) and trials, which were analyzed in the same manner as outlined for analyses of diagnosis. The third set of analyses combined data across all participants and examined conditioning effects by an ANOVAR model that included two factors: stimulus type (CS+, CS−) and trials, which were analyzed in the same manner as previously outlined.

The ANOVA results for the main effects and interactions of interest are presented in Table 3. Results for effects involving the trials factor are presented in the text. All significance levels reported for analyses that included the trials factor reflect the Greenhouse–Geisser correction for sphericity.

3.3.1. PTSD diagnosis effect

As can be seen in Table 3, there was no evidence of any differences between the PTSD and non-PTSD groups for CS interval SC responses during habituation, acquisition, or extinction. Furthermore, there were no significant diagnosis × trials (all Fs ≤ 1.5, ps $\geq .20$) or diagnosis × stimulus × trials (all Fs ≤ 1.5, ps $\geq .20$) interactions for any of the phases. Thus, acquisition and extinction of an aversively condition SC response did not differ between individuals with and without PTSD. The absence of significant diagnosis main effects also indicates that overall SC reactivity during the conditioning procedure did not differ between the PTSD and non-PTSD groups.

3.3.2. Effect of propranolol

Administration of propranolol did not influence the magnitude of SC responses or differential conditioned responses during habituation, acquisition, or extinction. As can be seen from Table 3, there were no significant condition main effects or condition × stimulus interactions during habituation, acquisition, or extinction. There also were no significant condition × trials (all Fs < 1) or condition × stimulus × trials (all Fs ≤ 1.1, ps $\geq .36$) interactions.

3.3.3. Combined data

The mean, averaged across all participants, SC response scores for the CS+ and CS− trials during habituation, acquisition, extinction, and retention are presented in Fig. 1A. The mean SC response scores for the UCS intervals of CS+ and CS− trials during acquisition are presented in Fig. 1B.

3.3.3.1. Habituation phase. As would be expected, and as can be seen from Fig. 1A and Table 3, SC response magnitudes did not differ between CS+ and CS− trials during the habituation phase. A marginally significant trials main effect ($F(4, 108) = 3.1$, $p = .06$) indicated that the SC response magnitude decreased over trials. The stimulus × trials interaction was not significant ($F(4, 108) = 1.8$, $p = .18$).

Table 3
ANOVA results for skin conductance (μS) CS interval responses

	d.f.s	Experimental phase									
		Habituation		Acquisition		Extinction		Non-threat retention		Threat retention	
		F	p	F	p	F	p	F	p	F	p
Stimulus	1, 27	1.2	.28	15.3	<.001	<1	n.s.	<1	n.s.	10.0	.004
Diagnosis	1, 26	<1	n.s.	<1	n.s.	<1	n.s.	1.3	.27	<1	n.s.
Diagnosis × stimulus	1, 26	<1	n.s.	<1	n.s.	<1	n.s.	2.7	.11	<1	n.s.
Condition	2, 25	<1	n.s.	<1	n.s.	<1	n.s.	<1	n.s.	1.4	.27
Condition × stimulus	2, 25	<1	n.s.	1.0	.37	<1	n.s.	1.1	.35	<1	n.s.

Note: CS = conditioned stimulus; stimulus effect = CS+ trials vs. CS− trials; diagnosis effect = PTSD vs. non-PTSD; condition effect = order in which placebo and propranolol were received, i.e., placebo/placebo vs. placebo/propranolol vs. propranolol/placebo; ANOVA = analysis of variance.

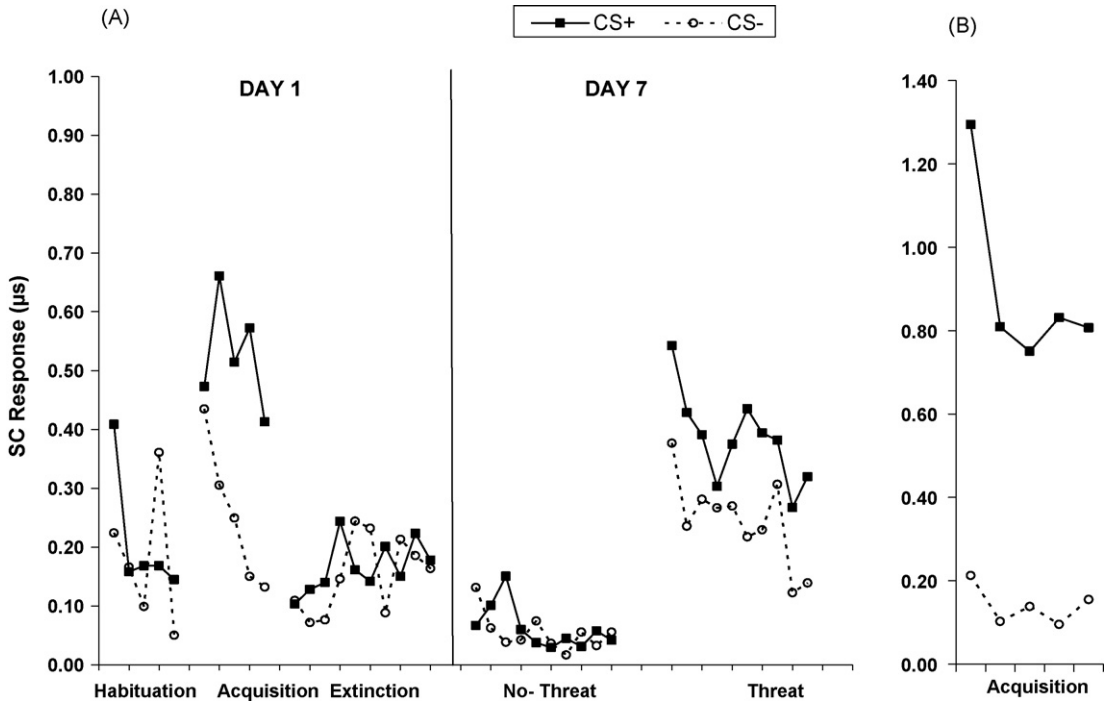

Fig. 1. Panel A depicts the group mean ($n = 28$) skin conductance (SC) response scores for the conditioned stimulus (CS) interval of CS+ and CS− trials during the habituation, acquisition, and extinction phases for day 1, and the no-threat and threat phases for day 7. Panel B depicts the group mean ($n = 28$) skin conductance (SC) response scores for the unconditioned stimulus (UCS) interval of CS+ and CS− trials during the acquisition phase (day 1).

3.3.3.2. Acquisition phase. Acquisition of a differentially conditioned SC response is evident in the significant stimulus main effect (Table 3). As can be seen in Fig. 1A, SC response magnitudes were larger to the reinforced CS+ trials compared to the non-reinforced CS− trials. Response magnitudes to the CS+ initially increased and then decreased over successive presentations, whereas response magnitudes to the CS− consistently decreased over trials. This pattern of reactivity produced a significant stimulus × trials interaction ($F(4, 108) = 3.5$, $p = .02$). The trials main effect was not significant ($F(4, 108) = 1.8$, $p = .15$).

Examination of Fig. 1B and the ANOVA results indicate that SC response magnitudes for the UCS interval of reinforced trials were significantly larger than those for the UCS intervals of non-reinforced trials (stimulus main effect, $F(1, 27) = 69.0$, $p < .001$). The SC response magnitudes for the UCS interval decreased over trials (trials main effect, $F(4, 108) = 5.7$, $p < .01$) and tended to do so at different rates for CS+ versus CS− trials (stimulus × trials interaction, $F(4, 108) = 3.1$, $p = .06$).

3.3.3.3. Extinction phase. There was no evidence of any differential SC response to CS+ versus CS− trials during the extinction phase. As can be seen from Fig. 1A, SC response magnitudes to previously reinforced and non-reinforced CSs were very comparable. The absence of a significant stimulus main effect (Table 3) and the absence of a trials main effect ($F(9, 243) < 1$) and stimulus × trials interaction ($F(9, 243) < 1$) indicate that the loss of differential responding to the CS+ versus CS− was apparent at the very outset of the extinction phase.

3.4. Retention procedure

3.4.1. PTSD diagnosis effect

As can be seen in Table 3, the PTSD and non-PTSD groups did not appear to differ in their differential SC responses to CS+ versus CS− non-threat and threat trials during retention. However, during the non-threat phase there was a trend towards initially *smaller* SC response magnitudes in the PTSD, compared to the non-PTSD, group as revealed by a marginally significant diagnosis × trials interaction ($F(9, 225) = 2.9$, $p = .06$). This tendency was not apparent during the threat phase (diagnosis × trials interaction, $F(9, 225) < 1$). The diagnosis × stimulus × trials interaction for the non-threat ($F(9, 225) = 1.8$, $p = .18$) and threat ($F(9, 225) < 1$) conditions did not approach statistical significance.

3.4.2. Effect of propranolol

Administration of propranolol during the previous laboratory session had no detectable effect on SC response magnitudes during the non-threat and threat conditions, as can be seen in Table 3 and in the absence of significant condition × trials (non-threat, $F(18, 216) < 1$; threat, $F(9, 216) = 1.3$, $p = .27$) or condition × stimulus × trials (non-threat, $F(18, 216) = 1.4$, $p = .24 < 1$; threat, $F(9, 216) = 1.2$, $p = .31$) interactions.

3.4.3. Combined data

The mean, averaged across all participants, SC response scores for the CS+ and CS− trials during non-threat and threat phases of the retention condition are presented in the right-hand portion of Fig. 1A.

3.4.4. Non-threat phase

There was no evidence of retention of a differential conditioned SC response to CS+ versus CS− trials during the non-threat condition. As can be seen from Fig. 1A, the SC response magnitudes for CS+ and CS− trials were comparable. The absence of a stimulus main effect (Table 3), trials main effect ($F(9, 234) = 1.3$, $p = .28$), and stimulus × trials interaction ($F(9, 234) = 1.5$, $p = .23$) indicates a lack of differential responding to CS+ and CS− that was evident at the beginning and across non-threat trials.

3.4.5. Threat phase

Examination of Fig. 1A and the ANOVA results presented in Table 3 reveal a pattern of significantly larger SC responses to CS+ versus CS− trials during the threat condition. Although there was a tendency for SC response magnitudes to become somewhat smaller over trials (trials main effect, $F(9, 234) = 2.0$, $p = .09$), the absence of a significant stimulus × trials interaction ($F(9, 234) < 1$) indicates that the differential response to the CS+ versus CS− trials did not get smaller. In order to determine whether the differential conditioned response differed from before to after the single UCS was administered following the fifth CS+ presentation, an additional analysis was performed that included time (before UCS, after UCS) as a factor in the AVOVA model. The absence of stimulus × time ($F(1, 29) < 1$) and stimulus × time × trials ($F(1, 29) < 1$) interactions indicates that the differential conditioned response before and after the UCS was presented did not significantly differ in magnitude.

3.5. Contextual conditioning

The effect of propranolol on contextual conditioning was examined by means of ANOVAR that included two factors: condition (propranolol, no propranolol), which was analyzed as a between-subjects effect, and phase, which served as the repeated measure. The phase factor contained three levels that corresponded to the mean 2 s, pre-stimulus SC levels measured during extinction trials of the first session and during the threat and non-threat trials of the second session. Because these SC level scores were non-normally distributed, the natural log of each score was used in the analyses. The log-transformed score means and S.D.s for the extinction, threat, and non-threat trials for the no propranolol condition are: $M = 1.35$, S.D. = .77; $M = .94$, S.D. = .73; $M = 1.10$, S.D. = .82, respectively, and for the combined propranolol conditions are: $M = 1.56$, S.D. = .91; $M = .86$, S.D. = .66; $M = 1.30$, S.D. = .76, respectively. Results of ANOVAR yielded a non-significant condition main effect ($F(1, 25) < 1$) and condition × phase interaction ($F(2, 50) = 1.1$, $p = .34$). These negative results suggest that propranolol had no discernable effect on contextual conditioning, as assessed from pre-stimulus SC levels.

There was a significant phase main effect ($F(2, 50) = 12.0$, $p < .001$), indicating that pre-stimulus SC levels differed over the extinction, non-threat, and threat phases. This effect was decomposed using the SAS profile option, which yielded significant differences between the mean SC levels for the extinction versus non-threat phases ($F(1, 25) = 18.2$, $p < .001$) and the non-threat versus threat phases ($F(1, 25) = 13.5$, $p = .001$).

3.6. Contingency awareness

Seven of 27 individuals (data from one individual were not available) failed to correctly identify the relationship between the CS+ and UCS. In order to assess whether this failure might have impaired acquisition and retention of a differential conditioned response, *t*-test comparisons between aware and non-aware subgroups were performed on differential SC response scores for acquisition phase and the threat phase of the retention condition. These experimental phases were chosen because they were associated with significant differences in SC response magnitudes to CS+ versus CS− trials. A differential conditioned response score was calculated for each phase by averaging SC responses to CS+ trials and subtracting the averaged SC response to CS− trials. The means, S.D.s, and results of *t*-test comparisons for the differential conditioned response scores for the aware and

Table 4
Pearson correlations between selected skin conductance variables

SC measures	1	2	3	4	5	6	7	8
1. Resting level	–	.43*	.58**	−.32	.51**	−.18	.82**	.40*
2. OR		–	.59**	.19	.26	−.08	.38†	.11
3. UCR			–	−.03	.35†	.09	.35†	.02
4. Habituation diff.				–	−.16	.05	−.32	.06
5. Acquisition diff.					–	−.25	.54**	.60**
6. Extinction diff.						–	−.51**	−.22**
7. Non-threat retention diff.							–	.34†
8. Threat retention diff.								–

Note: SC = skin conductance; resting level = mean during 5 min rest period; OR = orienting response, or averaged response to first presentation of the CS+ and CS− during the habituation phase; UCR = averaged unconditioned response for CS+ trials during the acquisition phase; diff. = differential; habituation, acquisition, extinction, non-threat and threat retention differentials = the averaged CS interval response to CS+ trials minus the averaged CS interval response to CS− trials for the habituation, acquisition, extinction, retention: non-threat and threat phases, respectively; CS = conditioned stimulus.

* $p < .05$.

** $p < .01$.

† $p < .10$.

non-aware subgroups are as follows, for the acquisition phase: aware, $M = .28$ μS, S.D. = .39; non-aware, $M = .28$ μS, S.D. = .35; $t(25) = .0$, $p = .99$; for the threat phase of retention: aware, $M = .16$ μS, S.D. = .20; non-aware, $M = .09$ μS, S.D. = .31; $t(24) = .7$, $p = .48$. Thus, inability to correctly specify the relationship between the CS+ and UCS did not appear to adversely influence acquisition and retention of a differential conditioned SC response.

3.7. Correlational analyses

Pearson correlations were used to examine relationships among the various indices of SC activity. As can be seen in Table 4, resting SC level showed significant positive relationships with mean SC orienting response (habituation phase), UCR, and the differential conditioned responses during acquisition and retention. The mean SC orienting response magnitude showed a positive correlation with the mean UCR. The differential conditioned response during the acquisition phase was positively correlated with the differential responses during the threat and non-threat phases of retention, but not during the extinction phase. The mean UCR showed a near significant positive correlation with the differential conditioned response during the acquisition phase.

4. Discussion

The present study found that: (1) administration of 40 mg of propranolol prior to cued conditioning had no effect on the acquisition of a differential conditioned SC response; (2) propranolol administered either prior to or following differential conditioning had no effect on the retention of extinction memory or reinstatement of conditioned fear. In addition, conditioned responses were found to be reinstated by threat-related cues (i.e., shock electrodes), thereby indicating that acquired conditioned responses remained durable for at least 1 week after conditioning. The heightened conditionability previously reported for individuals with PTSD (Orr et al., 2000) was not found in the current PTSD sample.

Findings from the present study clearly demonstrate that an aversively conditioned SC response is not eliminated by extinction training, and that the CR can be retained after a long delay. One week following aversive conditioning, there was no evidence of prior conditioning when participants knew there was no possibility of receiving the shock UCS. In the non-threat phase of the retention condition the differential response to CS+ versus CS− trials was near 0. However, the differential CR immediately reemerged when the shock electrodes were reattached to participants' fingers and they were informed that a shock might be presented. The lack of CR during the non-threat phase and the reinstatement of the CR during the threat phase indicate that both extinction and conditioning memories co-existed 1 week after conditioning and extinction training. These data extend further support to the hypothesis, first put forward by Pavlov (1927), that extinction does not erase the CS–UCS association (Quirk, 2002; Myers and Davis, 2002; Bouton, 2000).

It is important to note that the conditioning trials and subsequent retesting occurred in the same laboratory context. Because the differential CR was not evident during the non-threat phase of retention testing, i.e., when there was no veridical threat of shock, it is clear that the general laboratory cues did not provide a sufficiently threatening context for the CR to emerge. Only when the shock electrodes were reattached and information given about the possibility of receiving a shock was the CR reinstated. Thus, whereas CR responses in animals are easily reinstated by an unsignaled UCS (Bouton, 1993; Corcoran and Maren, 2001), here we demonstrate that presentation of threat-related cues alone were able to reinstate the CR.

Propranolol had no discernable effect on any phase of the cued differential conditioning procedure. This is consistent with previous animal and human studies. Although administration of propranolol has been found to reduce memory consolidation for contextual conditioning in animals (Roozendaal et al., 2004), several studies have shown that it had no effect on the memory consolidation of cued conditioning (Lee et al., 2001; Miserendino et al., 1990). As noted earlier, a study of human conditioning by Grillon et al. (2004) also did not observe an effect of propranolol on memory for cued conditioning. However, in contrast to the Grillon et al. findings, the present study observed no effect of propranolol on contextual conditioning, as measured from pre-stimulus SC levels during the non-threat and threat phases of retention testing. A possible reason for this discrepancy is suggested by the somewhat atypical pattern of SC level findings in the placebo group of the Grillon et al. study. Skin conductance levels typically decrease from first to second laboratory sessions, perhaps as individuals habituate to the novelty of laboratory testing. This reduction was observed in the placebo and propranolol groups of the present study, and in the propranolol group of the Grillon et al. study. However, the group that received placebo in the Grillon et al. study showed a slight increase in SC level from the first to second session. It seems likely that it is the placebo group's failure to show the expected reduction in SC level that is primarily responsible for the significant group difference, which served as the basis for Grillon et al.'s conclusion that propranolol influenced contextual conditioning.

Although the single 40 mg dose of propranolol used in the present study is the same as that used by Grillon et al., the observed effect on resting HR was relatively small. Whereas Grillon et al. observed a 7.7 BPM difference between the propranolol and placebo groups, in their HR change 1 h following drug ingestion, the present study only observed a 3.0 BPM difference over this same time interval. The decreased responsiveness to propranolol of our sample could be attributable to their older age, which was approximately 25 years older than the sample studied by Grillon et al. It may be that a 40 mg dose of propranolol is inadequate to produce measurable changes in the acquisition, extinction or retention of a conditioned response in older individuals. Thus, conclusions from the present study regarding the effect of propranolol on the acquisition, extinction and retention of a conditioned response must be tempered.

We were surprised by the failure to observe the heightened conditionability in PTSD patients that was so clearly evident in the previous study reported by our group (Orr et al., 2000). A recently published meta-analysis of studies that have examined classical fear conditioning in anxiety disorders has documented the somewhat weak and rather inconsistent findings across studies, especially those that have used a differential conditioning procedure (Lissek et al., 2005). As Lissek et al. note, stronger and more consistent group differences have been obtained from studies using a ''simple conditioning'' procedure, i.e., a single CS+. However, such studies have typically not controlled for non-associative effects such as sensitization. Consequently, it is not clear whether the differences observed between anxious and non-anxious groups during simple conditioning procedures reflect conditioning, sensitization or generally heightened reactivity. Lissek et al. also pointed out that some studies using differential conditioning have observed increased responding to the CS− as well as to the CS+ in anxious patient samples, raising the possibility that anxious individuals may have difficulty recognizing the CS− as a safety cue. Heightened reactivity to the CS− might contribute to a study's failure to observe group differences in differential conditioning, although this was not the case in our previous study (Orr et al., 2000) or even produce a finding of reduced differential responding in anxious patients. The failure to observe heightened differential conditioning in the present PTSD sample cannot be attributed to an increase in responding to the CS−. This would have been evident in a significant diagnosis main effect, as would be expected when there is increased responding to both the CS− and CS+, or a significant diagnosis × stimulus interaction, as would be expected if the differential response to the CS+ versus CS− was smaller in the PTSD group than in the non-PTSD group.

A few factors may have contributed to the failure to observe heightened conditionability in the PTSD sample. First, the PTSD sample of our previous study (Orr et al., 2000) was medication free, whereas only a small subset of the present PTSD sample was free from potentially confounding drugs or medications. It has become increasingly difficult to find medication-free veterans with PTSD, primarily due to the increasing age of Vietnam veterans. It is possible that medication use may have played a role in reducing anxiety or fearfulness, and thereby reduced the conditioning shown by some individuals with PTSD. However, in previous studies of startle reactivity (e.g., Metzger et al., 1999; Orr et al., 2003) and responses to trauma-related stressful materials (e.g., Orr et al., 1998) medication use has not been found to substantively influence psychophysiologic reactivity differences between PTSD and non-PTSD groups. Participants who were taking medications that could influence heart rate or blood pressure (e.g., alpha- or beta-blockers) were not excluded unless the medication was known to cross the blood–brain barrier (as does propranolol), but it seems unlikely that such medications could have directly or indirectly influenced conditionability.

A second possible reason for the failure to observe heightened conditionability in the PTSD sample is that the administration of the study ''medication'' (placebo or propranolol) 1 h prior to the start of testing may have served to reduce anxiety related to the conditioning procedure, especially in individuals with PTSD. Participants' belief that they might have received a drug that could reduce anxiety, or help their PTSD symptoms, may have served to reduce anxiety. A third possibility is that heightened conditionability is a feature of PTSD that is not shared by all individuals with the disorder, or demonstrated under all conditions. Consequently, it may be observed to varying degrees across PTSD samples and studies, as has been observed for other PTSD-related phenomena such as the eye-blink startle response (see Orr et al., 2004).

Acknowledgements

This project was supported by a grant from the Veterans Affairs Medical Research Service to Scott P. Orr. We wish to thank Michael L. Macklin and Heike B. Croteau for their technical assistance and the participants for their time and dedication.

References

Beck, A.T., Rush, A., Shaw, B.F., Emery, G., 1979. Cognitive Therapy of Depression. Guilford Press, New York.

Blake, D.D., Weathers, F.W., Nagy, L.M., Kaloupek, D.G., Gusman, F.D., Charney, D.S., Keane, T.M., 1995. The development of a clinician administered PTSD scale. Journal of Traumatic Stress 8, 75–90.

Bouton, M.E., 1993. Context, time, and memory retrieval in the interference paradigms of Pavlovian learning. Psycholological Bulletin 114, 80–99.

Bouton, M.E., 2000. A learning theory perspective on lapse, relapse, and the maintenance of behavior change. Health Psychology 19, 57–63.

Cahill, L., McGaugh, J.L., 1996. Modulation of memory storage. Current Opinion in Neurobiology 6, 237–242.

Cahill, L., Prins, B., Weber, M., McGaugh, J.L., 1994. Beta-adrenergic activation and memory for emotional events. Nature 371, 702–704.

Corcoran, K.A., Maren, S., 2001. Hippocampal inactivation disrupts contextual retrieval of fear memory after extinction. Journal of Neuroscience 21, 1720–1726.

Derogatis, L.R., 1983. SCL-90-R: Administration, Scoring, and Procedures Manual-II for the Revised Version. Clinical Psychometric Research, Towson, MD.

First, M.B., Spitzer, R.L., Gibbon, M., Williams, J.B.W., 1997. Structured Clinical Interview for DSM-IV Axis I Disorders (SCID-I), Clinician Version. American Psychiatric Press, Washington, DC.

Fowles, D.C., Christie, M.J., Edelberg, R., Grings, W.W., Lykken, D.T., Venables, P.H., 1981. Committee report. Publication recommendations for electrodermal measurements. Psychophysiology 18, 232–239.

Gould, T.J., 1998. Beta-adrenergic involvement in acquisition vs. extinction of a classically conditioned eye blink response in rabbits. Brain Research 780, 174–177.

Grillon, C., Cordova, J., Morgan III, C.A., Charney, D.S., Davis, M., 2004. Effects of the beta-blocker propranolol on cued and contextual fear conditioning in humans. Psychopharmacology 175, 342–352.

Harmer, C.J., Perrett, D.I., Cowen, P.J., Goodwin, G.M., 2001. Administration of the beta-adrenoceptor blocker propranolol impairs the processing of facial expressions of sadness. Psychopharmacology 154, 383–389.

Hatfield, T., McGaugh, J.L., 1999. Norepinephrine infused into the basolateral amygdala posttraining enhances retention in a spatial water maze task. Neurobiology of Learning and Memory 71, 232–239.

Introini-Collison, I., Saghafi, D., Novack, G.D., McGaugh, J.L., 1992. Memory-enhancing effects of post-training dipivefrin and epinephrine: involvement of peripheral and central adrenergic receptors. Brain Research 572, 81–86.

Ji, J.Z., Wang, X.M., Li, B.M., 2003a. Deficit in long-term contextual fear memory induced by blockade of beta-adrenoceptors in hippocampal CA1 region. European Journal of Neuroscience 17, 1947–1952.

Ji, J.Z., Zhang, X.H., Li, B.M., 2003b. Deficient spatial memory induced by blockade of beta-adrenoceptors in the hippocampal CA1 region. Behavioral Neuroscience 117, 1378–1384.

Lee, H.J., Berger, S.Y., Stiedl, O., Spiess, J., Kim, J.J., 2001. Post-training injections of catecholaminergic drugs do not modulate fear conditioning in rats and mice. Neuroscience Letters 303, 123–126.

Lissek, S., Powers, A.S., McClure, E.B., Phelps, E.A., Woldehawariat, G., Grillon, C., et al., 2005. Classical fear conditioning in the anxiety disorders: a meta-analysis. Behaviour Research and Therapy 43, 1391–1424.

McGaugh, J.L., Roozendaal, B., 2002. Role of adrenal stress hormones in forming lasting memories in the brain. Current Opinion in Neurobiology 12, 205–210.

Metzger, L.J., Orr, S.P., Ahern, C.E., Berry, N.J., Lasko, N.B., Pitman, R.K., 1999. Physiologic reactivity to startling tones in women with posttraumatic stress disorder. Journal of Abnormal Psychology 108, 347–352.

Miranda, M.I., LaLumiere, R.T., Buen, T.V., Bermudez-Rattoni, F., McGaugh, J.L., 2003. Blockade of noradrenergic receptors in the basolateral amygdala impairs taste memory. European Journal of Neuroscience 18, 2605–2610.

Miserendino, M.J., Sananes, C.B., Melia, K.R., Davis, M., 1990. Blocking of acquisition but not expression of conditioned fear-potentiated startle by NMDA antagonists in the amygdala. Nature 345, 716–718.

Myers, K.M., Davis, M., 2002. Behavioral and neural analysis of extinction. Neuron 36, 567–584.

Orr, S.P., Lasko, N.B., Metzger, L.J., Berry, N.J., Ahern, C.E., Pitman, R.K., 1998. Psychophysiologic assessment of women with posttraumatic stress disorder resulting from childhood sexual abuse. Journal of Consulting and Clinical Psychology 66, 906–913.

Orr, S.P., Metzger, L.J., Lasko, N.B., Macklin, M.L., Shalev, A.Y., Hu, F., Pitman, R.K., 2003. Psychophysiologic responses to sudden, loud tones in monozygotic twins discordant for combat exposure: relationship to posttraumatic stress disorder. Archives of General Psychiatry 60, 283–288.

Orr, S.P., Metzger, L.J., Lasko, N.B., Macklin, M.L., Peri, T., Pitman, R.K., 2000. De novo conditioning in trauma-exposed individuals with and without posttraumatic stress disorder. Journal of Abnormal Psychology 109, 290–298.

Orr, S.P., Metzger, L.J., Miller, M.W., Kaloupek, D.G., 2004. Psychophysiological assessment of PTSD. In: Wilson, J.P., Keane, T.M. (Eds.), Assessing Psychological Trauma and PTSD: A Handbook for Practitioners. 2nd ed. Guilford Press, New York, NY, pp. 473–542.

Orr, S.P., Pitman, R.K., Lasko, N.B., Herz, L.R., 1993. Psychophysiological assessment of posttraumatic stress disorder imagery in World War II and Korean combat veterans. Journal of Abnormal Psychology 102, 152–159.

Pavlov, I., 1927. Conditioned Reflexes. Oxford University Press, London.

Pitman, R.K., Sanders, K.M., Zusman, R.M., Healy, A.R., Cheema, F., Lasko, N.B., et al., 2002. Pilot study of secondary prevention of posttraumatic stress disorder with propranolol. Biological Psychiatry 51, 189–192.

Pitman, R.K., Shin, L.M., Rauch, S.L., 2001. Investigating the pathogenesis of posttraumatic stress disorder with neuroimaging. Journal of Clinical Psychiatry 62 (Suppl. 17), 47–54.

Quirk, G.J., 2002. Memory for extinction of conditioned fear is long-lasting and persists following spontaneous recovery. Learning and Memory 9, 402–407.

Roozendaal, B., 2002. Stress and memory: opposing effects of glucocorticoids on memory consolidation and memory retrieval. Neurobiology of Learning and Memory 78, 578–595.

Roozendaal, B., de Quervain, D.J., Schelling, G., McGaugh, J.L., 2004. A systemically administered beta-adrenoceptor antagonist blocks corticosterone-induced impairment of contextual memory retrieval in rats. Neurobiology of Learning and Memory 81, 150–154.

Salinas, J.A., Introini-Collison, I.B., Dalmaz, C., McGaugh, J.L., 1997. Post-training intraamygdala infusions of oxotremorine and propranolol modulate storage of memory for reductions in reward magnitude. Neurobiology of Learning and Memory 68, 51–59.

SAS Institute, 1982. SAS User's Guide: Statistics. SAS Institute, Cary, NC.

Spielberger, C.D., Gorsuch, R.L., Lushene, R.E., 1990. Manual for the State-trait Anxiety Inventory (Self-evaluation Questionnaire). Consulting Psychologists Press, Palo Alto, CA.

Sullivan, R.M., McGaugh, J.L., Leon, M., 1991. Norepinephrine-induced plasticity and one-trial olfactory learning in neonatal rats. Developmental Brain Research 60, 219–228.

Weiss, D.S., Marmar, C.R., 1997. The impact of event scale—revised. In: Wilson, J.P., Keane, T.M. (Eds.), Assessing Psychological Trauma and PTSD. Guilford Press, New York, pp. 399–411.