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Fear acquisition requires awareness in trace but not delay conditioning

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Abstract

The present study explored fear acquisition in differential delay versus trace conditioning in regard to the potential role of the acquired contingency awareness. One of two neutral pictures (CS+) either coterminated with (delay group; $n = 32$) or was followed by the aversive unconditioned stimulus (UCS) after CS offset (trace group; $n = 32$), while startle blink and skin conductance responses (SCR) were measured. As expected, the acquisition of conditioned startle potentiation in delay conditioning was independent of contingency awareness. In contrast, fear-potentiated startle in trace conditioning was only observed for those participants who were aware of the CS–UCS contingencies. SCR conditioning was generally only obtained for aware participants. The present results suggest a more implicit learning process in delay fear conditioning, whereas the explicit acquisition of contingency awareness might be a prerequisite for trace fear conditioning.

Descriptors: Fear, Startle, Delay conditioning, Trace conditioning, Awareness, Emotional learning

After being paired with an aversive event, previously innocuous stimuli become effective elicitors of a conditioned fear response. This classical (Pavlovian) fear conditioning is considered one of the most ubiquitous adaptive behaviors, because it enables the effective coping with potentially harmful stimuli and can be observed across a wide range of species including humans (cf. LeDoux, 1996). Moreover, classical fear conditioning is regarded as one common mechanism within the development of pathological states of fear and anxiety (e.g., Bouton, Mineka, & Barlow, 2001). Therefore, the experimental study of the mechanisms involved in the learning and unlearning of classically conditioned fear responses may promote important clinical implications (cf. Davis, 2002), which are based on the assumption that the similarity of the conditioning procedures in animal and human research expands to the learning mechanisms evoked by these procedures. In regard to the neural structures involved in fear conditioning, converging evidence from animal and human studies highlights the crucial role of the amygdala in regulating the acquisition, expression, and retention of a conditioned fear response (cf. Davis & Lang, 2003).

It must be noted, though, that humans participating in a typical fear conditioning study may not always acquire a genuine fear response, because the level of aversiveness of the unconditioned stimulus (UCS) is selected by the participant. Thus they may learn that the conditioned stimulus (CS) predicts the occurrence of the UCS without learning to *fear* the CS. In this case, participants acquire explicit declarative knowledge of the CS–UCS contingencies (cf. ‘‘contingency learning,’’ Rescorla, 1988; or ‘‘propositional learning,’’ Lovibond & Shanks, 2002), while the CS does not gain the *affective* properties to activate the fear system of the brain.

In support of this two-levels-of-learning account of fear conditioning (Öhman & Mineka, 2001), Hamm and Vaitl (1996) found a dissociation between different response systems in a classical conditioning experiment utilizing an aversive electric shock UCS and a nonaversive but significant UCS. This study revealed that conditioned skin conductance response (SCR) discrimination was observed for both aversive and nonaversive UCS conditions, whereas potentiation of the startle blink response was only observed during aversive fear conditioning. Moreover, potentiation of the startle eyeblink was obtained irrespective of the participants' declarative knowledge of the CS–UCS contingencies, whereas SCR conditioning was only reliable in those participants who acquired a cognitive representation of the contingencies.

These findings are clearly in line with the concept of multiple memory systems. Based on animal research and observations of amnesic patients, Squire and collaborators (Squire, 1992, 2004; Squire & Knowlton, 2000) developed a detailed taxonomy of different memory systems that involve a distinction between explicit, declarative memory systems (involving the conscious recollection of previous events) and implicit, nondeclarative memory systems, which are not expressed through conscious recollection but rather through performance (like skills, habits, or associative and nonassociative learning).

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One important line of evidence for the notion that multiple memory systems are involved in human classical conditioning comes from eyeblink classical conditioning, a prototype of motor learning based on cerebellar functioning. Eyeblink conditioning typically involves the presentation of an eyeblink-eliciting corneal air puff UCS and a preceding tone CS, which becomes an effective elicitor of the conditioned eyeblink response through a number of contingent CS–UCS pairings. A series of studies demonstrated that human eyeblink conditioning is obtained without declarative knowledge of the CS–UCS contingencies (cf. Clark & Squire, 1998). However, this only occurred using a delay conditioning procedure in which the UCS not only follows but also overlaps (and usually coterminates with) the CS presentation. In trace conditioning procedures, which involve a temporal gap between CS offset and UCS onset, the participants' awareness of the CS–UCS contingencies was a prerequisite for reliable eyeblink conditioning to occur (Clark & Squire, 1999). Further studies revealed that the differential impact of contingency awareness on eyeblink delay and trace conditioning is evident for both single-cue and differential conditioning procedures (Manns, Clark, & Squire, 2000a, 2000b, 2001; Smith, Clark, Manns, & Squire, 2005).

These data suggest that contingency awareness is not required if a motor response is learned in delay conditioning, but is a prerequisite for motor learning if there is a temporal gap between CS and UCS presentations. Given the findings about the implicit nature of fear acquisition described above, it is important to determine whether this dissociation between awareness, conditioning, and learning task also holds for *fear* learning. Surprisingly, to our knowledge there are only two studies so far that explicitly compared fear learning obtained in trace versus delay conditioning, and both studies observed pronounced conditioned SCR discrimination in both procedures (Hugdahl & Öhman, 1980; Lipp, Siddle, & Dall, 2003). On the other hand, Hugdahl and Öhman did not report on the participants' acquired awareness of the CS–UCS contingencies. Although Lipp et al. observed comparable amounts of conditioned fear-potentiated startle across the delay and trace conditioning groups, all but two participants demonstrated awareness of the CS–UCS contingencies, thus rendering an evaluation of the potential interaction between awareness, fear conditioning, and learning task difficult.

Two other studies, however, failed to obtain conditioned fear potentiated startle in delay conditioning in participants who were unaware of the CS–UCS contingencies (Grillon, 2002; Purkis & Lipp, 2001). In line with previous findings (Dawson & Schell, 1985; 1987; Hamm & Vaitl, 1996) both studies found that differential SCR conditioning was only observed for aware participants. However, in contrast to the results of Hamm and Vaitl (1996), neither study observed a conditioned fear potentiation of the startle response in unaware participants. These data are in line with single-process models of classical conditioning (cf. Lipp & Purkis, 2005; Lovibond & Shanks, 2002), which propose that—despite the existence of multiple memory systems—classical conditioning in *human* participants is based on propositional learning and thus is always closely tied to contingency awareness.

The discrepant findings of these studies and Hamm and Vaitl (1996) could be due to procedural differences. For example, the study by Purkis and Lipp (2001) involved a masking task as well as online measures of shock expectancy, which both might have affected the ongoing conditioning process, whereas Hamm and Vaitl (1996) utilized a standard differential conditioning design (like in animal experimentation) and included an independent postexperimental assessment of contingency awareness. Furthermore, in the study by Grillon (2002), raw blink magnitudes were analyzed, which are highly susceptible to individual outliers, whereas in the study by Hamm and Vaitl (1996), standardized scores were analyzed to ensure that each participant equally contributes to the group mean. Finally, whereas pictures of evolutionary significance (i.e., mutilations or erotic scenes) or human neutral faces served as conditioned stimuli in the study of Hamm and Vaitl, pictures of flowers and mushrooms or landscapes were utilized as CSs in the experiments of Grillon and Purkis and Lipp. A number of studies in preparedness research have revealed that fear conditioning is more difficult to obtain for the latter kinds of stimuli as compared to stimuli of evolutionary relevance (see Öhman & Mineka, 2001). Although conditioning effects are stronger if angry faces instead of neutral faces are used (Öhman & Dimberg, 1978), facial stimuli in general are significant for humans and thus might be associated more easily to motivationally relevant (unconditioned) stimuli than flowers or mushrooms.

Thus, in the present study we explored possible differences in fear acquisition between delay and trace paradigms depending on the acquired declarative knowledge of the CS–UCS contingencies. Based on previous research, the present study used a typical, differential conditioning design, that is, no information was provided about possible contingencies between the experimental stimuli. Two different pictures depicting neutral male facial expressions served as conditioned stimuli, and one of these was paired with the presentation of the aversive electric shock UCS during conditioning while startle blink and skin conductance responses were measured. The acquisition of declarative knowledge of the CS–UCS contingencies was evaluated in a final postexperimental interview in order not to interfere with the conditioning process.

Method

Participants

Sixty-four psychology students (49 women) of the University of Greifswald participated for course credit. Participants were randomly assigned to one of the two experimental groups, that is, 32 subjects (24 women) were assigned to the delay conditioning group ("delay") and 32 subjects (25 women) to the trace conditioning group ("trace"). The groups did not differ in mean age (delay: 21.9 years $\pm$ 0.59 *SE*; trace: 22.5 years $\pm$ 0.66 *SE*), $F(1,62) < 1$. All participants signed an informed consent form prior to the study, which was approved by the Ethics Committee of the University of Greifswald.

Stimulus Materials

Twelve pictures depicting male faces with neutral expressions were selected from the "Karolinska Directed Emotional Faces" (KDEF; Lundqvist, Flykt, & Öhman, 1998) to constitute a picture pool from which two pictures were individually selected for each participant to serve as conditioned stimuli. This selection was based on the participant's individual ranking of the pictures (see Procedure). The pictures were projected onto a screen approximately 2 m in front of the participants with a visible size of 55 $\times$ 85 cm. Picture duration was 6 s for the delay conditioning group and 2 s for the trace conditioning group, respectively. Picture presentation times were controlled by a tachistoscopic shutter (Gerbrands G1166), which was situated together with a slide projector (Kodak Ektapro 5000) in a room adjacent to the sound-shielded experimental room.

The unconditioned stimulus (UCS) comprised an electric stimulation (500 Hz monopolar DC-pulse) to the participant's left forearm in a 10-ms train of single pulses (1 ms) that was generated by a commercial stimulator (Grass Instruments S48K), isolated (SIU5) and transmitted via a constant current unit (CCU1) to a bipolar electrode (F-E10S2). The intensity of the UCS was individually adjusted to a level that was experienced as "highly annoying, but not painful." The mean physical intensity of the UCS was comparable between the delay and trace conditioning groups (delay: 8.8 mA $\pm$ 0.9 *SE*; trace: 9.6 mA $\pm$ 0.8 *SE*), $F(1,62) < 1$.

A 50-ms burst of white noise with an intensity of 95 dB[A] (rise/fall < 1 ms) served as the startle stimulus and was generated by a Coulbourn S81-02 and presented binaurally over headphones (Sony MDR-CD 170).

Physiological Recordings

Recordings of electromyographic (EMG) activity over the left orbicularis oculi muscle served to measure the eyeblink component of the startle response. Ag/AgCl miniature surface electrodes (Sensormedics) filled with electrolyte (Marquette Hellige) were attached beneath the lower eyelid using adhesive rings (Marquette Hellige). The raw EMG signal was amplified and filtered through a 30–1000-Hz bandpass, using a Coulbourn S75-01 bioamplifier. Digital sampling with a rate of 1000 Hz started 100 ms before until 400 ms after the onset of the acoustic startle stimulus. The EMG signal was filtered off-line through a 60-Hz highpass filter and was rectified and integrated (time constant: 10 ms) using a digital filter.

Skin conductance was recorded from the hypothenar eminence of the palmar surface of the participant's right hand. A Coulbourn S71-22 skin conductance coupler provided a constant 0.5 V across two Ag/AgCl standard electrodes (8 mm diameter; Marquette Hellige) filled with a 0.05 M sodium chloride electrolyte medium. The signal was processed with a resolution of 0.01 μS and sampled with a rate of 10 Hz. Data acquisition and stimulus presentations were synchronized using an IBM-compatible computer.

Procedure

Upon arrival at the laboratory subjects first read and signed an informed consent form. Then they were asked to rank 12 different picture cards of neutral male faces from the most to the least likeable picture. Six pictures (ranks 1, 2, 6, 7, 11, and 12) were selected for presentation during the preconditioning phase of the experiment. The two neither liked nor disliked faces (ranks 6 and 7) served as conditioned stimuli (CSs), that is, the rank 6 picture was dedicated as CS+ and the rank 7 picture as CS– for half of the subjects whereas this assignment was reversed for the other half of the subjects of each conditioning group.

The participants were seated in a reclining chair in an upright position and were told that a number of pictures would be presented and that sounds heard over the headphones could be ignored. After attaching the sensors for physiological data collection, six acoustic startle probes were presented for an initial habituation of the startle eyeblink to a stable baseline. Following these initial startle probes, the series of six neutral faces was presented with both to-be-conditioned stimuli presented last to serve as a baseline measure prior to conditioning. Both of the CSs and two other picture presentations were followed by an acoustic startle probe either 4.5 or 5.0 s after picture onset. Two further startle probes were presented during the intertrial intervals (ITI), which varied between 14 and 21 s.

After this preconditioning phase the experimenter entered the room and attached the electrodes for electric stimulation to the participant's left forearm. The intensity of the UCS was individually adjusted within five warned presentations to a level that was experienced as "highly annoying, but not painful." Participants were then instructed that during the following phase of the experiment, a series of pictures and some electric pulses would be presented; no information regarding the contingencies between the pictures and the electric pulses was provided.

The differential conditioning phase consisted of nine presentations of each CS, that is, one male neutral face (CS+) was always followed by the electric UCS, whereas the other male neutral face (CS–) was never paired with the UCS. The order of presentation of the CSs was randomized with the restriction of no more than two consecutive presentations of the same picture. Four different stimulus orders were created and the participants in each conditioning group were equally assigned to one of these orders. For the delay conditioning group, the CS+ always coterminated with the presentation of the UCS (CS–UCS onset asynchrony: 5990 ms). For the trace conditioning group the CS–UCS onset asynchrony was kept constant (5990 ms), thus resulting in 3990 ms between CS offset and UCS onset (see Figure 1). In six of nine presentations of each CS, an acoustic startle probe was administered either 4500 or 5000 ms after CS onset (three times each). Moreover, six acoustic startle probes were presented during the ITIs that varied between 14 and 22 s.

The conditioning phase was immediately followed by the postconditioning trials, that is, no signal indicated that no further UCSs would be presented. The postconditioning phase consisted of nine presentations of each CS. The order of CS presentation and the timing of the startle probes was the same as during the previous conditioning phase.

After the postconditioning phase, a postexperimental interview consisting of six questions was carried out to determine whether the participants were aware of the CS–UCS contingencies (cf. Bechara et al., 1995; Dawson & Schell, 1987). The interview started with a "free recall" question ("Did you know when

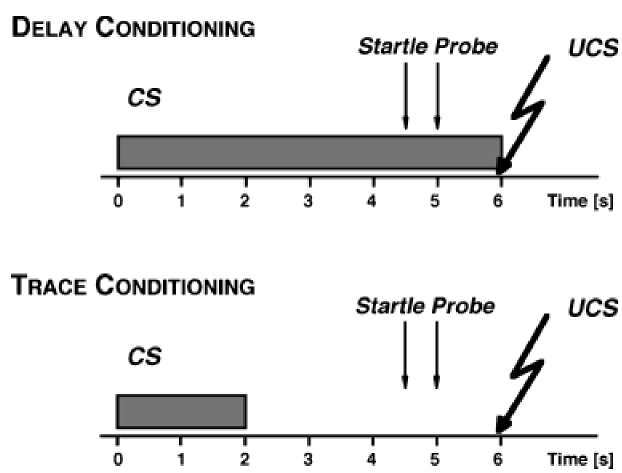

Figure 1. Experimental timing of CS and UCS presentations during the delay (upper panel) and the trace conditioning experiment (lower panel), respectively. Acoustic startle probes followed CS onset by either 4.5 or 5 s in two-thirds of the trials.

you were going to receive an electro-tactile stimulus?''). Then the participant was asked how many different faces he/she had seen during the experiment and a short description of each of the faces was requested. Three additional questions addressed possible differences between the faces (''Did you notice any differences between the pictures?'') and whether there might have been a relation between the presentation of the faces and the electro-tactile stimulus (''How many *different* pictures were followed by the electro-tactile stimulus?'' and ''Please name the picture or pictures that were followed by the electro-tactile stimulus.''). Finally, the interview ended with a multiple choice ''recognition'' question: ''The presentation of an electro-tactile stimulus followed: (1) the male face with . . . (participant's individual description provided by question #2), (2) the male face with . . . (participant's individual description provided by question #2), (3) there was no systematic relationship, or (4) not possible to answer.''

Data Reduction and Response Definition

Startle blink magnitudes. The magnitude of the startle eyeblink was scored off-line using a computer program (Globisch, Hamm, Schneider, & Vaitl, 1993) that identified latency of blink onset in milliseconds and peak amplitude in microvolts. Only blinks starting 20–100 ms after probe onset and peaking within 150 ms were scored. No detectable eyeblinks were scored as zero responses. Trials with excessive baseline activity or recording artifacts were rejected. Thirty-two out of 3072 trials (1.04%) had to be discarded. The number of rejected trials varied between zero and four trials per subject and the conditioning groups did not differ in number of missing trials (delay: 12 trials; trace: 20 trials; $F[1,62] = 1.26$, $p = .265$). Prior to the statistical data analyses, all missing values were replaced individually for each subject by the overall mean blink response magnitude of this subject over all 48 trials. Distribution analyses suggested that the startle blink magnitude data should be normalized. The standardized responses of each participant were converted to T-scores $[50+(z \times 10)]$.

Skin conductance responses. Skin conductance responses to the conditioned stimuli were scored as the largest increase in conductance between 0.9 and 4.0 s after stimulus onset (first interval response [FIR]; Prokasy & Kumpfer, 1973) using a computer program (Globisch et al., 1993). Skin conductance responses elicited by picture onset were unaffected by the acoustic startle probes, which were presented either 4.5 or 5.0 s after picture onset. The unconditioned responses were scored as the largest increase in conductance between 0.9 and 4.0 s after the onset of the electrical stimulus. Unconditioned responses did not differ between the delay and trace conditioning group (delay: $0.80\ \mu S \pm 0.12\ SE$; trace: $0.72\ \mu S \pm 0.11\ SE$; $F[1,62] < 1$).

Trials in which no response could be detected or with a response magnitude $< 0.05\ \mu S$ were considered as zero responses. Trials with respiration or recording artifacts were rejected (105 out of 3584 trials; 2.93%). The number of discarded trials did not exceed seven trials per subject and did not differ for the two conditioning groups (delay: 64 trials; trace: 41 trials). Missing values were replaced individually for each participant by the mean magnitude for all picture trials (averaged across CS+ and CS– presentations) or the mean unconditioned response magnitude averaged across all UCS presentations. Logarithms of all values were computed to normalize the distribution (Venables & Christie, 1980). To reduce interindividual variability that was not related to the conditioning task, the log values were range corrected by dividing each individual score by the participant's maximum response within all CS and UCS trials (Lykken & Venables, 1971).

Awareness of the CS–UCS contingencies. The protocols of the postexperimental interviews were thoroughly evaluated independent from the evaluation of the physiological response data. Fifty percent of the participants ($N = 32$ of 64; delay: $n = 15$, trace: $n = 17$) were able to correctly name the CS–UCS contingencies in the initial free recall question. However, 16 additional participants (delay: $n = 9$, trace: $n = 7$) were able to correctly identify the CS+ throughout the further interview. Although 6 of these participants stated in the final multiple-choice question that there was ''no systematic relationship'' ($n = 4$) or that this question was ''not possible to answer'' ($n = 2$) they had been able nonetheless to correctly report that only one picture (the CS+) was followed by the electro-tactile stimulus (questions #4 and #5). Therefore, these participants were classified as aware in order to avoid ''false negatives'' in considering participants as unaware. Thus, overall 16 participants (25%), 8 subjects within each conditioning group, failed to show any declarative knowledge of the CS–UCS contingencies during the entire interview. These participants either chose ''no systematic relationship'' ($n = 13$) or ''not possible to answer'' ($n = 3$) in the final multiple-choice question and stated throughout the entire interview that both pictures were followed by the electro-tactile stimulus. Therefore, these participants were classified as ''unaware.''

As illustrated in Table 1 neither the UCS intensity nor the magnitude of the unconditioned skin conductance response differed between aware and unaware participants either in delay or in trace conditioning, $Fs < 1$. These data suggest that the aversive UCS was equally effective in aware and unaware participants.

Data Analysis

The different phases of the experiment were analyzed separately. For the analysis of response sensitization due to the repeated presentation of the aversive UCS during the work-up procedure, responses to the first CS presentation during conditioning were compared to the last CS presentation in the preconditioning phase, irrespective of whether these trials comprised a CS+ or a CS–. Mixed-model ANOVAs were calculated for the startle

Table 1. *Physical Intensity of the Unconditioned Stimulus (UCS) and the Unconditioned Skin Conductance Response Magnitudes for the Participants, Who Were either Aware or Unaware of the CS–UCS Contingencies in Differential Delay or Trace Conditioning, Respectively*[a]

	Delay conditioning		Trace conditioning	
	Aware (n = 24)	Unaware (n = 8)	Aware (n = 24)	Unaware (n = 8)
Shock (UCS) (mA)	8.3 (0.8)	10.2 (2.6)	10.1 (1.0)	8.3 (1.2)
Skin conductance response (μS)	0.88 (0.14)	0.55 (0.15)	0.75 (0.14)	0.63 (0.15)

[a]Mean values (SE) are provided.

blink and skin conductance responses with between-subjects factors of Group (delay vs. trace) and Awareness (aware vs. unaware) and within-subjects factors of Conditioning (CS+ vs. CS– for skin conductance; CS+ vs. CS– vs. ITI for startle blink responses) and Trial Block (first [trials 1–3] vs. second [trials 4–6] vs. third [trials 7–9]). Degrees of freedom were corrected where appropriate using Greenhouse–Geisser's ε. Uncorrected degrees of freedom are reported along with the according ε value. Partial η^2 values are provided as a measure of effect size.

Results

Preconditioning

Startle blink magnitudes. Prior to conditioning, startle responses that were elicited during (delay) or after (trace) the presentation of the to-be-conditioned stimuli ($M_{\text{CS}+} = 46.91$, $SE = \pm 1.17$; $M_{\text{CS}-} = 46.39$, $SE = \pm 0.99$) tended to be smaller than the startle responses elicited during the ITI ($M_{\text{ITI}} = 50.43$, $SE = \pm 1.23$), $F(2,120) = 2.69$, $p = .077$, $\varepsilon = .91$, $\eta^2 = .04$. Startle response magnitudes did not differ for the designated CS+ and CS– prior to conditioning, $F < 1$. No differences were observed between conditioning groups or between aware and unaware participants, $Fs < 1$.

Skin conductance responses. Skin conductance responses to the designated CS+ and CS– did not differ prior to conditioning, $F(1,60) = 1.08$, $p = .303$. Again, no differences were observed between the delay and the trace conditioning group or between aware and unaware participants, $Fs < 1$.

Sensitization following UCS Exposure

Startle blink magnitudes. Startle blink responses during (delay) or after (trace) the first CS presentation following the UCS workup were significantly larger than those elicited during or after the last CS onset prior to the workup, $F(1,60) = 19.73$, $p < .001$, $\eta^2 = .25$ ($M_{\text{pre}} = 47.31$, $SE = \pm 1.08$; $M_{\text{post}} = 58.79$, $SE = \pm 1.41$). This pronounced sensitization of the startle reflex after repeated electric shock exposure did not differ between both conditioning groups, $F(1,60) = 2.46$, $p = .121$, or between aware and unaware participants, $F(1,60) = 2.59$, $p = .113$. Moreover, no interaction between Group and Awareness was observed for the sensitization of the startle response magnitudes, $F < 1$.

Skin conductance responses. As was the case for startle blink magnitudes, UCS exposure resulted in a pronounced increase in skin conductance magnitude to the first CS presentation in the conditioning phase, $F(1,60) = 32.22$, $p < .001$, $\eta^2 = .35$ ($M_{\text{pre}} = 0.05$, $SE = \pm 0.01$; $M_{\text{post}} = 0.21$, $SE = \pm 0.02$). Again, this sensitization was equally pronounced in both conditioning groups and also across aware and unaware participants, $Fs < 1$.

Conditioning

Startle blink magnitudes. As expected, startle responses elicited during (delay) or after (trace) the presentation of the CS+ were significantly potentiated compared to those elicited during the ITI, $F(1,60) = 13.00$, $p < .01$, $\eta^2 = .18$, and those elicited during or after the presentation of CS–, $F(1,60) = 6.44$, $p < .05$, $\eta^2 = .10$, resulting in a main effect of Conditioning, $F(2,120) = 8.63$, $p < .01$, $\varepsilon = .89$, $\eta^2 = .13$. As reflected in the nonsignificant interaction of Conditioning $\times$ Group, $F < 1$, conditioned startle modulation was equally pronounced in both delay conditioning, $F(2,60) = 5.84$, $p < .01$, $\varepsilon = .91$, $\eta^2 = .16$, and

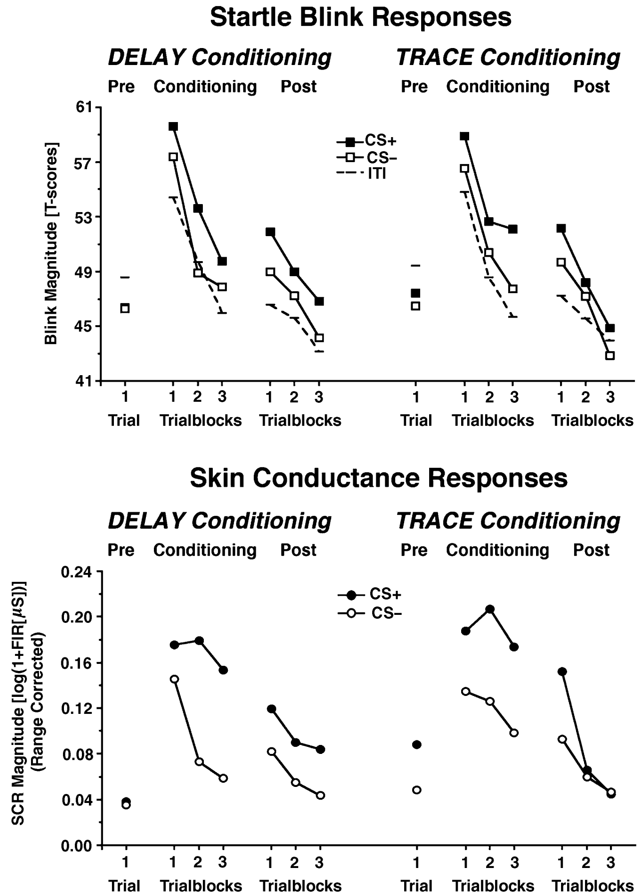

Figure 2. Mean startle blink (upper panel) and skin conductance (lower panel) response magnitudes in differential delay and trace fear conditioning. Startle responses were elicited by an acoustic probe during (delay) or after (trace) the CS+ or CS– presentations, or during the intertrial interval (ITI), respectively. Single trials are illustrated during the preconditioning phase (Pre), whereas two trials (startle blink responses) or three trials (skin conductance responses) were blocked during the conditioning and the postconditioning (Post) phases.

trace conditioning (see Figure 2, upper panel), although the main effect of conditioning was marginal in the latter group, $F(2,60) = 2.97$, $p = .067$, $\varepsilon = .86$, $\eta^2 = .09$.

Interestingly, the conditioned startle modulation in the trace conditioning group was further qualified by a significant interaction with the participants' awareness, Conditioning $\times$ Awareness $F(2,60) = 6.16$, $p < .01$, $\eta^2 = .17$, indicating robust conditioning in the aware subjects, $F(2,46) = 18.67$, $p < .001$, $\varepsilon = .78$, $\eta^2 = .45$, whereas no conditioned startle modulation could be observed in the unaware participants of the trace conditioning group, $F < 1$. Startle blink responses of the aware participants in the trace conditioning group were potentiated when elicited after the presentation of the CS+ compared to both ITI, $F(1,23) = 28.57$, $p < .001$, $\eta^2 = .55$, and CS–, $F(1,23) = 11.10$, $p < .01$, $\eta^2 = .33$, whereas neither conditioned startle potentiation nor discrimination could be observed in the unaware participants of the trace conditioning group, both $Fs < 1$.

In contrast, the robust conditioned startle modulation observed in delay conditioning was *not* further modulated by the participants' awareness, Conditioning $\times$ Awareness $F < 1$. During delay conditioning, a pronounced conditioned startle modulation was observed in aware participants, $F(2,46) = 7.46$,

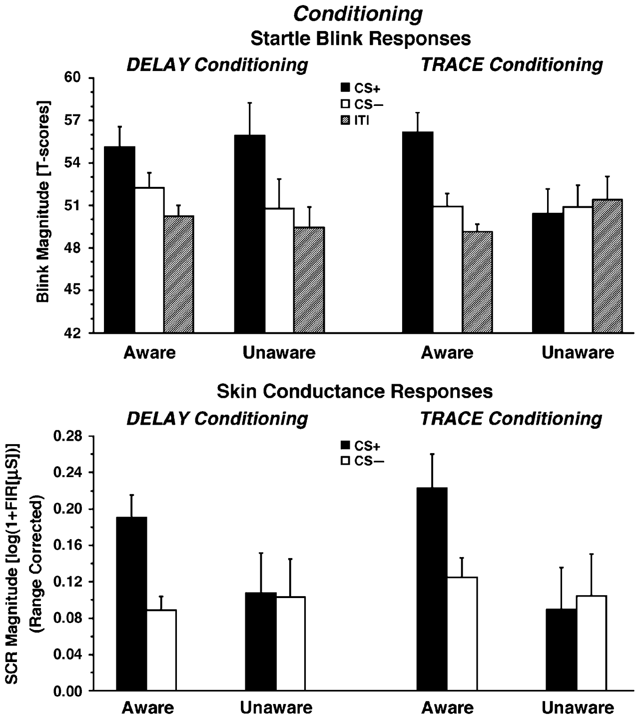

Figure 3. Mean startle blink (upper panel) and skin conductance (lower panel) response magnitudes during the conditioning phase in differential delay and trace fear conditioning, for participants, who were either aware (each $n = 24$) or unaware (each $n = 8$) of the CS–UCS contingencies. Startle blink responses elicited at later probe times (probes presented 990 ms preceding the UCS onset) are illustrated.

$p < .01$, $\varepsilon = .90$, $\eta^2 = .25$, whereas the main effect of Conditioning fell short of statistical significance for the unaware participants, $F(2,14) = 1.46$, $p = .267$, $\varepsilon = .84$, $\eta^2 = .17$. However, further analyses revealed that conditioned startle modulation varied for the two different startle probe times in the unaware participants in the delay conditioning group, Conditioning × Probe Time $F(2,14) = 4.86$, $p < .05$, $\varepsilon = .75$, $\eta^2 = .41$. Startle blink responses elicited 990 ms before the onset of the UCS were substantially potentiated in the eight unaware participants[1] both relative to the ITI, $F(1,7) = 4.48$, $p = .072$, $\eta^2 = .39$, and to the CS–, $F(1,7) = 5.05$, $p = .059$, $\eta^2 = .42$, whereas no substantial startle potentiation or discrimination could be observed for the earlier startle probe time (1490 ms before UCS onset), both $Fs < 1$. Aware participants of the delay conditioning exhibited significant startle potentiation and discrimination, $Fs(1,23) = 11.30$ and 6.97, $p < .05$, η^2s = .33 and .23, irrespective of the probe time, $F < 1$. Moreover, the different probe times did not affect conditioned startle modulation during trace conditioning, $Fs < 1$. The upper panel of Figure 3 depicts the startle blink magnitudes for the second probe time for aware and unaware participants of the delay and trace conditioning (see Appendix for both probe times' trial by trial responses).

Skin conductance responses. As expected, skin conductance responses to the CS+ were significantly larger than those to the CS–, $F(1,60) = 10.26$, $p < .01$, $\eta^2 = .15$ (see Figure 2, lower panel). Replicating previous findings, this conditioned discrimination was modulated by the participants' contingency awareness, $F(1,60) = 12.67$, $p < .01$, $\eta^2 = .17$. As illustrated in the lower panel of Figure 3, only those participants who correctly recognized the CS–UCS contingencies in the postexperimental interview exhibited larger skin conductance responses to the CS+ compared to the CS– during both delay and trace conditioning, $Fs(1,23) = 28.63$ and 13.48, $p < .001$, η^2s = .56 and .37. In contrast, unaware participants failed to show any significant differential electrodermal conditioning in either procedure, $Fs < 1$, resulting in a significant interaction of Conditioning × Awareness, $Fs(1,30) = 7.92$ and 5.56, $ps < .01$ and .05, η^2s = .21 and .16 for delay and trace conditioning, respectively. Thus, in contrast to the conditioned startle blink responses, skin conductance conditioning was generally only observed for aware subjects, irrespective of whether a trace or a delay conditioning procedure was used.[2]

Relationship between startle blink response and skin conductance. Correlational analyses were conducted to evaluate the relationship between the different measures of fear conditioning within subjects. The magnitude of conditioned startle potentiation (CS+ minus ITI), startle discrimination (CS+ minus CS–), and conditioned skin conductance discrimination (CS+ minus CS–) during the conditioning phase of the experiment were assessed for each participant, and correlations between these different indices of fear conditioning were calculated.

In the delay conditioning group, neither conditioned startle potentiation nor conditioned startle discrimination were related to the amount of conditioned discrimination in skin conductance, $r = .14$ and .18, $p > .325$. In contrast, in the trace conditioning group, the conditioned electrodermal response discrimination was clearly related to the observed startle potentiation, $r = .47$, $p < .01$, and startle discrimination, $r = .54$, $p < .01$. These relationships between the amount of conditioned startle modulation and skin conductance conditioning were also observed for the aware participants in the trace conditioning group, $r = .43$ and .51, $p < .01$ (skin conductance discrimination correlated with startle potentiation and discrimination, respectively), demonstrating that the covariation between these conditioning measures during trace conditioning were not artificially induced by the subgroup of unaware participants.

Postconditioning

Startle blink magnitudes. Startle response magnitudes elicited during (delay) or after (trace) the CS+ continued to be potentiated in the postconditioning phase as reflected by a significant main effect of Conditioning, $F(2,120) = 4.54$, $p < .05$, $\varepsilon = .98$, $\eta^2 = .07$ (see Figure 2, upper panel). Follow-up tests revealed larger startle blink responses following CS+ onset relative to the ITI, $F(1,60) = 8.59$, $p < .01$, $\eta^2 = .13$, but not to the CS–, $F(1,60) = 1.85$, $p = .179$. However, unlike during the conditioning phase, conditioned startle modulation was generally more pronounced in aware compared to the unaware participants, Conditioning × Awareness, $F(2,120) = 4.43$, $p < .05$, $\eta^2 = .07$, irrespective of the conditioning group, $F(2,120) = 1.64$, $p = .199$.

[1]The main effect of conditioned startle modulation, $F(2,14) = 3.28$, $p = .075$, $\varepsilon = .90$, $\eta^2 = .32$, falls short of statistical significance if raw scores are entered in the analysis, $F(2,14) = 2.35$, $p = .163$, $\varepsilon = .57$, $\eta^2 = .25$, demonstrating the importance of using standardized scores, especially in small samples.

[2]Skin conductance results were not further modulated by trial type; that is, these effects were observed irrespective of which kinds of CS trials (either with or without acoustic early or late startle probes) were entered in the analyses.

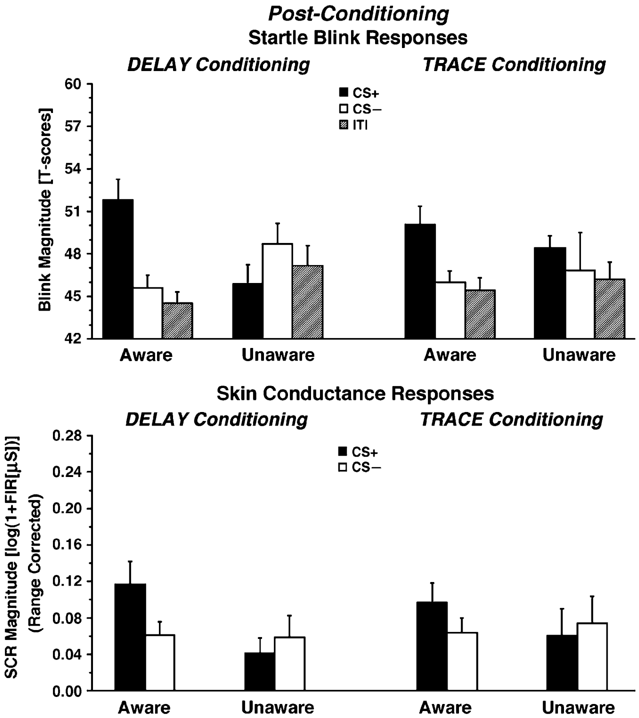

Figure 4. Mean startle blink (upper panel) and skin conductance (lower panel) response magnitudes during the postconditioning phase in differential delay and trace fear conditioning for participants, who were either aware (each $n = 24$) or unaware (each $n = 8$) of the CS–UCS contingencies. Startle blink responses elicited at later probe times (probes preceding the former UCS onset by 990 ms) are illustrated.

As illustrated in Figure 4 (upper panel), the aware participants continued to demonstrate robust conditioned potentiation of the startle blink response in the delay conditioning group, $F(2,46) = 11.00$, $p < .001$, $\varepsilon = .87$, $\eta^2 = .32$, as well as in the trace conditioning group, $F(2,46) = 5.10$, $p < .05$, $\varepsilon = .96$, $\eta^2 = .18$. Similar to the conditioning phase of the experiment, the unaware participants of the trace conditioning group exhibited no conditioned startle modulation, $F < 1$. However, unlike during the conditioning phase, the unaware participants of the delay conditioning group also failed to show substantial conditioned startle modulation during the postconditioning phase, $F(2,14) = 1.08$, $p = .343$. Further analyses revealed that during the postconditioning phase, the two different startle probe times did not further modulate startle response magnitudes of the unaware participants in either the delay or trace conditioning groups, $Fs(2,14) < 1$, whereas both startle potentiation and discrimination were more pronounced for the later startle probe time in the aware participants of the delay and trace conditioning groups, $Fs(1,46) = 9.45$ and 7.41, $p < .01$, η^2s = .17 and .14, respectively.

Skin conductance responses. Similar to the conditioning phase of the experiment, skin conductance responses to the CS+ were larger than those to the CS– only in those participants who were aware of the CS–UCS contingencies, which was reflected by a significant Conditioning × Awareness interaction, $F(1,60) = 5.91$, $p < .05$, $\eta^2 = .09$. As illustrated in Figure 4 (lower panel), conditioned skin conductance discrimination was maintained during the postconditioning in the aware participants of both conditioning groups, $Fs(1,23) = 5.97$ and 5. 56, $p < .05$, η^2s = .21 and .20 for delay and trace conditioning, respectively, whereas it was absent for the unaware participants in the delay and trace conditioning group, $Fs < 1$. Moreover, the conditioned skin conductance discrimination observed for the aware participants during the postconditioning phase was more readily extinguished in the trace conditioning group, as indicated by the interaction of Conditioning and Trial Block, $F(2,46) = 3.43$, $p < .05$, $\varepsilon = .96$, $\eta^2 = .13$, compared to the delay conditioning group, demonstrating robust differential skin conductance responding throughout the postconditioning, Conditioning × Trial Block $F < 1$.

Relationship between startle blink response and skin conductance. Similar to the conditioning phase, robust covariations between the magnitude of conditioned startle modulation and skin conductance discrimination were observed in the aware participants of the trace conditioning group, $r = .44$ and .46, $p < .05$ (skin conductance discrimination correlated with startle potentiation and discrimination[3]), whereas no such correlations could be observed for the aware participants of the delay conditioning group, $r < .27$, $p > .202$, thus further substantiating the independence of both measures in delay fear conditioning.

Discussion

The present study explored the mediation of fear response acquisition in differential delay and trace conditioning by declarative knowledge of the stimulus contingencies assessed in a postexperimental interview. As expected, startle blink responses were markedly potentiated when elicited during the shock-reinforced CS during delay conditioning and also when probes were delivered a few seconds after the offset of the CS+ in trace conditioning. Thus, replicating previous findings (Lipp et al., 2003), the acquisition of a conditioned startle response potentiation was comparably pronounced during delay and trace conditioning.

The current results also clearly indicate that conditioning performance in delay and trace conditioning was differentially mediated by the participants' declarative knowledge of the stimulus contingencies. Such an analysis was not possible in previous studies (e.g., Lipp et al., 2003), because most participants exhibited contingency awareness, possibly promoted by explicitly instructing participants to attend to the CS *and* UCS presentations. In contrast, in the current study the participants' attention was not selectively drawn to the UCS presentations, and 25% of the participants were not able to subsequently report the stimulus contingencies, despite the use of rather conservative criteria for assessing declarative knowledge.

Replicating previous findings (Hamm & Vaitl, 1996), the acquisition of conditioned startle potentiation to the reinforced CS was not modulated by contingency awareness in delay conditioning. This result supports numerous data suggesting that fear responses can be acquired by implicit learning without necessarily requiring the explicit knowledge of the contingencies (see Öhman & Mineka, 2001). On the other hand, the present study also found a clear dissociation between conditioned startle

[3]Because conditioned startle potentiation and discrimination effects were more pronounced for the second, compared to the first, startle probe time during postconditioning, the correlational analyses focused on the amount of conditioned startle modulation observed for the second startle probe time.

potentiation and the required learning task. Whereas implicit learning of startle potentiation was obtained in delay conditioning, no conditioned startle potentiation was found for participants without contingency awareness in the trace conditioning task.

Thus, the present data are amazingly comparable to the findings obtained for motor learning. In particular, previous studies on eyeblink classical conditioning have repeatedly demonstrated the same dissociation between contingency awareness and conditioning task on the acquisition of conditioned responses. In a delay conditioning task, the conditioned eyeblink response to a tone CS predicting a corneal air puff UCS is independent of the declarative knowledge of the CS–UCS contingencies, whereas in a trace conditioning task such propositional learning is a prerequisite for eyeblink conditioning to occur (cf. Clark & Squire, 1999). Accordingly, amnesic patients with hippocampal damage show clear impairments in trace eyeblink conditioning but exhibit normal performance in a delay conditioning procedure (Clark & Squire, 1998), suggesting that the hippocampus might be important for trace conditioning and contingency awareness.

While the present study revealed a clear dissociation of startle fear potentiation and contingency awareness depending upon the learning task, a different pattern of results was observed for skin conductance conditioning. Replicating the findings of Hugdahl and Öhman (1980), robust skin conductance conditioning was found in both conditioning tasks. In contrast to conditioned startle potentiation, a close association between electrodermal conditioning and contingency awareness was observed that was independent of the learning task. That is, substantial conditioned skin conductance discrimination only occurred in those participants who were able to verbally report the CS–UCS contingencies, irrespective of whether a delay or a trace conditioning procedure was employed. Although cue-specific potentiation of the startle blink response was observed in the unaware participants of the delay conditioning group, no conditioned skin conductance discrimination was observed in this group.

Although affective modulation of the startle response is especially pronounced for highly arousing aversive pictures that also elicit larger skin conductance responses (Cuthbert, Bradley, & Lang, 1996), these data suggest that increased electrodermal responding does not seem to be necessary for startle potentiation to occur. For example, reliably potentiated startle during viewing of unpleasant pictures is observed even when skin conductance responses have habituated due to repeated presentations of the unpleasant stimuli (Bradley, Cuthbert, & Lang, 1993). Moreover, pronounced fear potentiated startle in the absence of cue-specific skin conductance responses to the fear-eliciting, visual stimulus has recently been observed in a patient with bilateral cortical blindness (Hamm et al., 2003), further substantiating the notion that the emotional impact of a stimulus is not completely reflected by increases in skin conductance.

It should be noted, however, that the differential impact of contingency knowledge on the conditioned startle blink and skin conductance measures in delay, as compared to trace, conditioning was especially pronounced *during* the conditioning phase and could not be observed in the postconditioning phase of the experiment. Specifically, the conditioned startle potentiation that was observed for the unaware participants in the delay conditioning group was readily extinguished during the postconditioning phase, suggesting that the acquired fear potentiation was less robust. Moreover, the conditioned startle potentiation of the unaware participants in the delay conditioning group was specifically observed for startle eliciting probes that preceded the occurrence of the UCS more closely. Although the probe delays did not differ by more than 500 ms, the later probe time matched the interstimulus interval (ISI) between the CS and UCS more closely. Such a temporal specificity of the fear potentiated startle is a well-known finding in animal research; that is, the potentiation of the startle response is more pronounced for those startle eliciting probes that are presented at testing intervals that match the CS–UCS interval used in conditioning training. This temporal specificity is observed even with a single conditioning training trial (Davis, Schlesinger, & Sorenson, 1989) and is independent of whether a trace or delay conditioning procedure is utilized (Burman & Gewirtz, 2004). Thus, the temporal specificity of the potentiated startle responses observed in the present study lends further support to the validity of the current findings.

Focusing on the conditioning phase of the experiment, the present study revealed a clear dissociation of conditioned fear-potentiated startle and skin conductance discrimination for delay conditioning procedures. In contrast, no such dissociation could be observed for the trace conditioning task, suggesting that the acquisition of a fear response by means of classical conditioning might involve different mechanisms when delay or trace conditioning procedures are carried out. Importantly, this notion was further substantiated by the correlational analyses. Specifically, in the delay fear conditioning group, the actual amount of either conditioned startle potentiation or discrimination was unrelated to the amount of conditioned skin conductance discrimination, whereas these measures closely covaried in trace conditioning. Moreover, this close coherence between the startle and skin conductance conditioning measures in trace conditioning, as well as their independence in delay conditioning, were observed during both the conditioning and the postconditioning phase of the experiment. This is especially noteworthy because the between-group analyses suggested a more similar pattern of conditioned responding for delay and trace procedures.

Taken together, the present results suggest that when the UCS follows the CS after an empty interval, as in trace conditioning, the acquisition of declarative knowledge might be a prerequisite of learning (irrespective of the response system analyzed), whereas in delay conditioning, when the UCS and CS overlap, the acquisition of a fear response (as indexed by a potentiation of the startle blink responses) does not depend on explicit awareness. Such conclusions are in line with models proposing that these learning tasks might access different neural circuits. According to the broad evidence that declarative memory is based on the functional integrity of the hippocampal formation (cf. Squire, Stark, & Clark, 2004), animal research has revealed that trace (as opposed to delay) fear conditioning is a hippocampus-dependent task (for an overview see Fanselow & Poulos, 2005). Moreover, recent brain imaging studies in humans lend further support for the involvement of the hippocampus in trace fear conditioning (Büchel, Dolan, Armony, & Friston, 1999; Knight, Cheng, Smith, Stein, & Helmstetter, 2004). Furthermore, distraction from the conditioned stimuli interferes with fear conditioning in trace but not delay conditioning in both mice (Han et al., 2003) and men (McKell Carter, Hofstötter, Tsuchiya, & Koch, 2003), indicating that attention appears as a prerequisite for trace (but not delay) fear conditioning.

In regard to the neural underpinnings of implicit (emotional) and explicit (declarative) learning, a large body of evidence from animal and human research consistently indicates that the acquisition, expression, and retention of a fear response requires intact amygdala functioning (cf. Davis, 2000; Fanselow, 1994;

Koch, 1999; LeDoux, 1996; Maren, 2001). However, whether the acquisition of declarative knowledge about stimulus contingencies is similarly closely related to hippocampal functioning still needs to be critically evaluated, especially in light of the structural and functional complexity of the hippocampal formation and adjacent structures of the temporal lobe (for an overview, see Squire et al., 2004). Thus far, the functional integrity of the hippocampal formation appears as a prerequisite for declarative knowledge to develop, but likely there is more to declarative knowledge than hippocampal activity. In support of this notion, a recent brain imaging study revealed hippocampal activity in both aware and unaware participants in a sensory learning paradigm (McIntosh, Rajah, & Lobaugh, 2003). Interestingly, the differences between aware and unaware participants involved the functional connectivity between hippocampal and other (especially frontal) brain areas; that is, the unaware participants showed spatially more restricted network activations, which did not expand to frontal areas.

As a caveat, it should be mentioned that Bechara et al. (1995) observed complete blocking of electrodermal conditioning with retained declarative knowledge of the CS–UCS contingencies in a patient with a confined bilateral amygdala lesion and intact electrodermal conditioning in the absence of declarative knowledge of CS–UCS contingencies in a patient with bilateral hippocampal damage, thus confirming the dual process model of conditioning but also qualifying the close relationship between electrodermal conditioning and contingency awareness. Contrary to the results of most group studies, which are in line with the present findings, Bechara et al. (1995) report a single patient with extensive, repeated conditioning training. Although skin conductance conditioning might be more closely related to emotional arousal under these circumstances, skin conductance responses to the CS+ observed in group studies involving a single conditioning session might index emotional arousal or orienting as well. At the least, the close relationship between electrodermal conditioning and contingency awareness observed in the present and previous group studies suggests that conditioned skin conductance discrimination might primarily index increased orienting to the reinforced CS.

The present study conclusively reveals that declarative knowledge of the CS–UCS contingencies appears to have a differential impact on fear acquisition in differential delay, as compared to trace, conditioning. Although fear acquisition as indexed by the conditioned potentiation of the startle response was observed in the absence of contingency awareness in delay conditioning, no aversive learning without awareness was found for trace conditioning. Moreover, skin conductance conditioning was specifically observed for aware (but not for unaware) participants irrespective of the conditioning procedure. Thus, electrodermal conditioning seems to primarily index cognitive learning of the rules or circumstances in which a specific stimulus is signaling an aversive event, which is a declarative and explicit memory.

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APPENDIX

Table A1 shows the mean (*SE*) startle blink and skin conductance response magnitudes of the aware and unaware participants of the delay and trace conditioning.

Table A1. *Mean (SE) Startle Blink and Skin Conductance Response (SCR) Magnitudes of the Aware (AW) and Unaware (UNAW) Participants of the Delay (DEL) and Trace (TRC) Conditioning*[a]

		Startle blink responses					SCR	
		Probe time 1		Probe Time 2				
		CS+	CS–	CS+	CS–	ITI	CS+	CS–
DEL-AW (*n* = 24)								
Preconditioning[b]	1	47.3 (2.7)	47.3 (2.2)	46.5 (2.2)	44.5 (2.4)	49.5 (1.3)	.03 (.01)	.04 (.02)
Conditioning	1	59.7 (1.6)	57.5 (1.8)	61.1 (2.2)	60.4 (2.1)	54.9 (1.8)	.19 (.03)	.14 (.02)
	2	51.0 (1.5)	45.7 (1.2)	56.1 (3.0)	50.1 (1.5)	49.9 (1.2)	.20 (.03)	.07 (.02)
	3	51.9 (1.9)	48.2 (1.6)	48.2 (1.6)	46.3 (1.6)	45.9 (0.8)	.18 (.04)	.06 (.02)
Postconditioning	1	51.2 (1.5)	49.7 (1.7)	54.0 (2.5)	47.0 (1.5)	45.6 (1.4)	.14 (.03)	.07 (.02)
	2	48.7 (1.8)	46.0 (2.2)	51.1 (1.9)	45.0 (1.6)	44.9 (0.9)	.11 (.03)	.05 (.02)
	3	46.1 (2.2)	44.3 (1.5)	50.2 (1.6)	44.6 (1.5)	43.0 (1.2)	.10 (.03)	.06 (.02)
DEL-UNAW (*n* = 8)								
Preconditioning[b]	1	43.1 (2.8)	49.9 (3.4)	51.0 (6.3)	45.8 (2.3)	45.8 (1.0)	.05 (.04)	.02 (.02)
Conditioning	1	54.8 (3.0)	52.3 (2.9)	59.4 (4.6)	53.4 (3.8)	53.0 (2.7)	.14 (.05)	.15 (.06)
	2	52.9 (3.3)	54.2 (2.4)	54.7 (3.0)	49.6 (2.3)	49.1 (2.2)	.11 (.05)	.09 (.04)
	3	44.0 (2.3)	50.3 (3.4)	53.7 (5.2)	49.4 (2.5)	46.2 (1.8)	.07 (.03)	.07 (.03)
Postconditioning	1	48.3 (2.7)	48.7 (2.5)	51.4 (1.8)	53.3 (2.7)	49.8 (1.6)	.06 (.03)	.11 (.04)
	2	47.2 (1.1)	57.3 (5.3)	45.5 (2.5)	47.6 (2.6)	47.9 (2.8)	.02 (.01)	.06 (.03)
	3	45.0 (3.1)	41.4 (4.0)	40.7 (2.2)	45.0 (2.1)	43.7 (4.0)	.05 (.02)	.01 (.01)
TRC-AW (*n* = 24)								
Preconditioning[b]	1	44.7 (2.5)	44.6 (3.4)	47.9 (3.4)	46.7 (1.9)	48.5 (1.4)	.10 (.03)	.05 (.02)
Conditioning	1	62.1 (2.1)	59.3 (1.4)	59.7 (2.2)	54.7 (1.9)	54.4 (1.1)	.22 (.03)	.14 (.02)
	2	53.8 (2.2)	51.1 (3.0)	53.5 (2.5)	50.6 (1.6)	48.3 (1.2)	.25 (.04)	.14 (.02)
	3	51.8 (1.6)	47.5 (1.7)	55.2 (2.4)	47.5 (1.1)	44.6 (1.2)	.21 (.05)	.10 (.03)
Postconditioning	1	49.8 (1.7)	51.7 (2.0)	55.3 (2.0)	49.9 (1.8)	47.3 (1.3)	.18 (.04)	.10 (.02)
	2	47.5 (1.7)	47.2 (1.9)	48.3 (1.3)	45.1 (1.4)	45.6 (1.4)	.07 (.02)	.05 (.01)
	3	44.9 (1.5)	42.7 (1.1)	46.5 (1.9)	42.8 (1.2)	43.3 (1.4)	.04 (.02)	.05 (.02)

Table A1. *(Continued)*

		Startle blink responses					SCR	
		Probe time 1		Probe Time 2				
		CS+	CS–	CS+	CS–	ITI	CS+	CS–
TRC-UNAW ($n = 8$)								
Preconditioning[b]	1	41.3 (3.1)	53.1 (3.0)	61.2 (4.8)	44.4 (2.5)	52.2 (3.6)	.04 (.02)	.04 (.03)
Conditioning	1	51.6 (2.6)	50.6 (2.1)	54.0 (2.8)	59.5 (4.0)	56.0 (3.5)	.10 (.04)	.13 (.05)
	2	52.5 (5.3)	51.1 (2.9)	47.0 (2.7)	47.1 (3.2)	49.5 (3.0)	.09 (.05)	.09 (.05)
	3	45.7 (3.2)	50.9 (3.4)	50.2 (1.2)	46.1 (3.5)	48.7 (1.7)	.08 (.05)	.09 (.05)
Postconditioning	1	49.6 (2.9)	47.5 (3.3)	52.7 (1.6)	45.5 (1.5)	47.1 (1.7)	.07 (.05)	.08 (.04)
	2	46.9 (2.2)	49.1 (4.6)	51.0 (3.2)	51.5 (4.8)	45.6 (2.1)	.06 (.03)	.10 (.05)
	3	43.4 (2.0)	43.0 (3.5)	41.5 (2.2)	43.4 (2.4)	45.8 (2.6)	.04 (.03)	.04 (.02)

[a]SCRs were elicited by the onset of the conditioned stimuli (CS), whereas startle blinks were elicited by acoustic startle probes presented either 4500 ms (probe time 1) or 5000 ms (probe time 2) after CS onset or during the intertrial interval (ITI). Standardized scores are provided (startle blink [T scores]; SCR [log(1+FIR [μS])], range-corrected).

[b]Note that sample sizes are smaller for CS related startle blink responses during the preconditioning phase, because each CS presentation was accompanied by either the first or the second startle probe time.