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One-trial overshadowing: Evidence for fast specific fear learning in humans

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ABSTRACT

Adaptive defensive actions necessitate a fear learning system that is both fast and specific. Fast learning serves to minimize the number of threat confrontations, while specific learning ensures that the acquired fears are tied to threat-relevant cues only. In Pavlovian fear conditioning, fear acquisition is typically studied via *repetitive* pairings of a *single* cue with an aversive experience, which is not optimal for the examination of fast specific fear learning. In this study, we adopted the one-trial overshadowing procedure from basic learning research, in which a combination of two visual cues is presented once and paired with an aversive electrical stimulation. Using on-line shock expectancy ratings, skin conductance reactivity and startle reflex modulation as indices of fear learning, we found evidence of strong fear after a single conditioning trial (fast learning) as well as attenuated fear responding when only half of the trained stimulus combination was presented (specific learning). Moreover, specificity of fear responding tended to correlate with levels of state and trait anxiety. These results suggest that one-trial overshadowing can be used as a model to study fast specific fear learning in humans and individual differences therein.

1. Introduction

Fear is the ensemble of emotional reactions that is triggered when danger is imminent. It serves to motivate defensive actions that cope with the upcoming dangerous event. While some fears are genetically pre-programmed, most fears are acquired through Pavlovian conditioning (Davey, 1992; Lissek et al., 2005). This consists of learning experiences in which dangerous encounters (unconditional stimulus, US) become associated with preceding innocuous cues (conditional stimulus, CS) that henceforth signal the US and elicit fear (conditional reaction). In the natural environment, fear learning is adaptive when it is *fast* (in order to minimize confrontations with dangerous events) and *specific* to the actual situation that carries the danger potential.

Fast specific fear learning is challenged by the fact that danger situations are often complex in nature and involve multiple preceding cues (i.e., potential CSs). Failures in fast specific learning under these circumstances may contribute to fear overgeneralization patterns as typically seen in anxiety-related disorders. Post-traumatic stress disorder, for example, is characterized by psychological and physiological distress during exposures to subsets of cues that resemble aspects of the traumatic event (American Psychiatric Association, 2013; Ehlers & Clark, 2000; Ehlers et al., 2002). However, the challenge of fast specific fear learning is not readily captured in the prototypical fear conditioning procedure where a *single* stimulus (CS) is paired *multiple* times with an aversive stimulus (US). The purpose of the current study was to develop a pre-clinical human fear conditioning protocol for the study of fast specific fear learning, given its relevance for adaptive and clinical forms of anxiety.

The standard fear conditioning procedure has been adjusted before to study either fast or specific learning. First, studies showed that humans (Öhman, Eriksson, & Olofsson, 1975) and non-human animals (Fanselow, 1990; Jarvik & Essman, 1960) can learn to fear simple neutral stimuli and even more complex contexts following a *single* learning trial (fast learning). Second, *specificity* has been

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studied via tests of fear generalization between simple stimuli that vary on a single perceptual dimension (Dymond, Dunsmoor, Vervliet, Roche, & Hermans, 2015). For example, fear conditioning to a circle is followed by generalization tests with a range of circles of varying sizes (Lissek et al., 2008). This typically results in an orderly descending fear gradient as a function of perceptual similarity. Context generalization research extended this research to more complex situations that simultaneously differ on multiple perceptual dimensions (Rudy & O'Reilly, 1999; Rudy & O'Reilly, 2001). Hence, fear generalization research has focused mainly on the effects of gradual changes in conditioned stimuli and contexts. Much less is known, however, about the degree to which fear conditioned to a stimulus complex generalizes to its constituent stimuli when encountered separately. This lacuna is surprising; because conditions like PTSD are markedly characterized by inflated fear reactions to isolated trauma cues *as if* the whole traumatic situation is once again present. In sum, while procedures have been developed to examine either fast or specific fear learning, little is known about the combined challenge of fast specific fear learning in complex stimulus situations.

Basic associative learning research in non-human animals has provided a simple, yet elegant procedure that lends itself perfectly to the study of fast specific learning in complex stimulus situations. One-trial overshadowing (OTO, a variant of the multiple-trial overshadowing procedure; Kaufman & Bolles, 1981; Lanzetta & Orr, 1980, 1981; Mackintosh, 1976; Pavlov, 1927) refers to a procedure in which a combination of two CSs is presented once, followed by the US. Subsequently, the CSs are presented separately and the degree of conditioned responding is measured. Under these conditions, non-human animals show lower conditioned responding compared to a CS that was individually paired with the US (Cole, Oberling, & Miller, 1999; James & Wanger, 1980; Kaye, Gambini, & Mackintosh, 1988; McNally & Westbrook, 2003; Mackintosh, 1971; Mackintosh & Reese, 1979; but see; Zelikowsky & Fanselow, 2010). According to standard associative learning theory, this reduction reflects *configural processing* of the CS complex during the conditioning trial (see Kaye et al., 1988). With stronger configural processing, the fear association is thought to accrue to the stimulus complex as a whole, rather than to its constituent stimuli. Separated presentations of these stimuli will therefore fail to activate the fear association and trigger little fear (the OTO-effect). According to non-associative theories, on the other hand, the fear decrease may primarily reflect the *uncertainty/ambiguity* that surrounds the first confrontation with part of a conditioned stimulus compound (probabilistic/propositional theories of conditioning, De Houwer, Vandorpe, & Beckers, 2005; Mitchell, De Houwer, & Lovibond, 2009). Interestingly, anxiety-related conditions like PTSD have been linked both to configural processing deficits (e.g., Gilbertson et al., 2007) and threat biases during situations of uncertainty/ambiguity (e.g., Beckers, Krypotos, Boddez, Effting, & Kindt, 2013; Lissek, Pine, & Grillon, 2006). These deficits/biases are expected to impair OTO. First, configural processing deficits may lead to the individual elements present during conditioning all acquiring an association with the US, resulting in excessive fear responding in OTO (see General Discussion). Second, threat biases would lead to an over-estimation of the threat value of the uncertainty/ambiguity surrounding OTO and trigger aberrant fear responding as well. Thus, OTO is both theoretically and clinically relevant to the study of anxiety.

The present study was set up to adapt and validate OTO as a model for fast specific fear learning in humans. A compound CS (AX) was paired with an aversive electrical stimulation once, followed by test presentations of one of the CSs (X). In addition, a single CS (B) was paired with the stimulation once, while another single CS (C) was not. Fast learning would be reflected by stronger fear reactions to B than to C (B > C). Selective learning would be reflected by lower fear reactions to X than to B (B > X). We also assessed state and trait levels of anxiety to explore influences of anxiety on fast selective fear learning in humans.

2. Experiment 1

2.1. Material and methods

2.1.1. Participants

Twenty psychology students (nine women) with a mean age of 23.4 (SD = 6.38) participated in experiment 1 in return for course credits. All participants gave informed consent and were aware that they could abort the experiment at any time.

2.1.2. Apparatus

2.1.2.1. Conditioned stimuli. Four geometrical shapes (trapezium, diamond, hexagon, cross) served as conditional stimuli and were presented on a computer screen (Dell LCD monitor, type 1708 FPc). These shapes were grey with a black border and presented in a white frame. Stimuli were presented against a black background screen.

2.1.2.2. Unconditioned stimuli. A 2-ms electrocutaneous stimulus administered to the wrist of the dominant hand served as a US. A Digitimer DS71 constant current stimulator (Hertfodshire, UK) administered the stimulus via a pair of V91-01-8-mm reusable Bilaney Ag/AgCL electrodes. Electrodes were filled with K-Y Jelly.

2.1.3. Measures

2.1.3.1. Questionnaires. Participants completed the Trait Anxiety inventory (STAI-T; Spielberger, 1983; Dutch translation by van der Ploeg, Defares, & Spielberger, 1981).

2.1.3.2. Skin conductance reactivity. Electrodermal activity was recorded using a skin conductance coupler manufactured by Coulbourn Instruments (model V71-23, Allentown, PA). A constant voltage of 0.5 V was applied across a pair of 8-mm Ag/AgCl electrodes. These electrodes were attached to the palm of the non-dominant hand. A Labmaster DMA 12 bit analog-to-digital converter (Scientific Solutions, Solon, Ohio) digitized the skin conductance signal at 10 Hz from 2 s prior to CS onset until 6 s after CS offset.

2.1.3.3. Shock-expectancy. To measure trial-by-trial subjective shock expectancy ratings, an eleven-point scale was used. This scale ranged from 0 to 10 and was labeled: “certainly no shock” (0), “maybe” (5), “certain shock” (10). A left mouse click on the scale registered the corresponding position for that trial.

2.1.4. Procedure

After participants gave their informed consent, they completed the STAI-T questionnaire. Next, electrodes were fitted and the shock intensity was set to a level that was determined “definitely uncomfortable, but not painful” through a standard shock work-up procedure. Subsequently, participants were instructed that pictures of geometrical shapes would appear on the computer screen and that some of these shapes could be followed by a shock. It was further explained that the participant's task was to predict the occurrence of the shock. Next, participants were instructed how to use the expectancy ratings scale.

The experiment consisted of two phases (see Table 1 and Fig. 1). During acquisition two stimuli (A and X) were presented in compound (AX) and followed by an electrical shock. Two additional stimuli (B and C) were each presented individually. Stimulus B was