Observed but Never Experienced – Vicarious Learning of Fear Under Ecological Conditions

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Abstract

Learning to avoid threats often occurs through indirect experiences, for example, by observing the behavior of others. Most previous research on observational learning has used pre-recorded stimuli to induce learning. Here, we aimed to enhance the ecological validity of the learning situation by inviting two friends to serve as the 'observer' and 'demonstrator', respectively. The observer watched their friend performing a differential fear-conditioning task in real time. During the task, one conditioned stimulus (CS+) was associated with an electric shock (unconditioned stimulus, US) to the forearm of the demonstrator. Another stimulus (CS-) was always safe. Following the learning phase, the observer was presented with the CS+ and CS-, but without receiving any shocks. As an index of learning, we measured their skin conductance and fear-potentiated startle responses. While the US applied to the demonstrator robustly elicited strong skin conductance responses in the observers during the learning phase, subsequent differential skin conductance responses of the observers (to CSs presented directly) were dependent on declarative knowledge of the CS+/US contingency. Contingency-aware observers also showed elevated fear-potentiated startle responses during both CS+ and CS- compared to intertrial intervals. Learning efficiency was lower than in previous studies. We conclude that observational fear learning involves two separable components: an automatic, non-specific emotional reaction to the response of the demonstrator (serving as a social US) and learning to predict stimulus contingency (CS+/US pairing). Ecological modifications proposed in the article mirror analogous rodent studies on vicarious fear conditioning.

Keywords: observational fear learning, ecological validity, fear-potentiated startle, contingency learning, fear conditioning

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– Vicarious Learning of Fear Under Ecological Conditions

Learning through interactions with others, i.e., social learning, is often adaptive. For instance, receiving information about threats through social means helps to avoid costly first-hand experiences. Social transfer of information about threats is thought to be mediated by emotional contagion, a bottom-up process through which the emotional state of one organism elicits the same state in another (Preston & de Waal, 2002). The basic ability to perceive the emotions of others is conserved across species. Rodents are able to learn about threats vicariously (Knapska et al., 2006, 2010). In humans, it has been shown that watching an actor undergoing fear conditioning evokes vicarious reactions in the observer and results in their learning as measured by behavioral, psychophysiological, and neural responses (Lindström et al., 2018; Olsson & Phelps, 2004). These responses were modulated by a number of factors, including the observer's empathy level (Olsson et al., 2016), social group affiliation (Golkar & Olsson, 2017), and racial similarity to the actor (Golkar et al., 2015).

The procedure used in the previous studies on vicarious fear learning involved presenting subjects with video recordings of an actor subjected to a differential fear conditioning task. After the observational fear learning phase (OFL), participants were asked to perform 'the same' task on their own (direct expression phase, DE). Crucially, no aversive stimulation was administered to the observers to ensure that they learned through indirect, social means only. Successful fear conditioning was indexed as an enhanced skin conductance response (SCR) to the conditioned stimulus, CS, which was paired with an electric shock administered to the actor during the OFL phase (the so called CS+). A protocol describing guidelines for human observational fear learning research has recently been published by Haaker et al. (2017). While this protocol is well designed for studying vicarious fear learning under well-controlled laboratory conditions, it does not take into account

experiments which might be performed in more natural situations, something which is critical in research focusing on social interactions (Bottenhorn et al., 2019; Matusz et al., 2019).

In this study we aimed to improve the ecological validity of the Haaker et al.'s protocol. Ecologically valid (naturalistic) paradigms are defined as involving realistic, interactive stimuli representative of real-world experiences while maintaining a reasonable degree of experimental control (Bottenhorn et al., 2019). We believe that modifying the procedure toward a more naturalistic one can tell us how observational fear learning occurs when authentic emotions are being expressed. For the purpose of adapting the protocol, we decided to invite pairs of participants and involve them both in the experimental procedure. Instead of using a prerecorded video of an actor, one of the two participants was asked to become a live demonstrator. Additionally, in order to create a more naturalistic situation, we recruited pairs of friends, considering the evolutionary value of learning from highly familiar others. It has also been suggested that ingroup members' feelings are highly informative and often used for learning about emotional valence of things (Kavanagh & Winkielman, 2016). Moreover, it has been shown that interpersonal liking increases emotional mimicry (McIntosh, 2006), which is yet another factor involved in emotional contagion (Olszanowski et al., 2019), and that learning from an ingroup demonstrator is more efficient compared to an outgroup one (Golkar & Olsson, 2017). Besides the idea of investigating genuine emotions in an authentic social context, the changes proposed here are one step in narrowing the translational gap between the human observational fear conditioning paradigm and contemporary experiments on rodents upon which the human experiments were based. We hope that this can be used as a starting point for further studies comparing behavioral and neural correlates between rodent and human models of vicarious fear acquisition.

In our study, one of the participants (the demonstrator) was subjected to a classical fear conditioning task while being watched by another participant (the observer) through a

live video stream. The skin conductance and fear-potentiated startle (FPS) responses of the observers were recorded as indicators of vicarious fear learning. Although both have been commonly used in aversive conditioning studies, FPS has only recently been applied in the observational fear conditioning design (Selbing & Olsson, 2019) and no studies from this field have so far employed both these measures, making our study the first to incorporate both. Their combination enables measuring the cognitive (SCR) and affective (FPS) components of the fear conditioning process.

Our hypotheses were as follows: first, we expected an augmented skin conductance response in the observers watching their friends receiving electric shocks during the OFL phase. Second, we hypothesized that as a consequence of a pairing between CS+ and social US (friend's reaction to the shock) watched during the learning phase, the observers would develop a conditioned response to the CS+ without directly experiencing the aversive stimulation. This socially driven conditioned reaction was expected to be reflected in skin conductance and fear-potentiated startle responses, both presumed to be stronger after CS+ compared to CS- presentation, when tested in the DE phase.

Methods

Participants

70 male volunteers (35 pairs of friends), aged between 18 and 27 (M = 21.4, SD = 2.2) participated in the study. The sample size was not determined through power analysis because the experiment was intended to establish procedure validity and sample size required for a planned fMRI study. To be eligible for the study, a pair had to have known each other for at least 3 years (in the recruited group: M = 6.8, SD = 4.2) and score sufficiently high (minimum 30 out of 60 points; in the recruited group: M = 50.1, SD = 7.3) on the McGill Friendship Questionnaire (Mendelson & Aboud, 1999). All the participants were screened for the ability to recognize the colors used in the task. Only heterosexual participants (based on

self-declaration) were included. Within each pair, the subjects were randomly assigned roles – one person was the demonstrator (learning model) and the other one was the observer. The study was approved by the local Research Ethics Committee at Faculty of Psychology, University of Warsaw. All subjects gave their written informed consents and received financial compensation for participation in the study.

Stimuli and Materials

Two large colored squares (blue and yellow) displayed on a computer screen in front of the learning model (demonstrator) served as conditioned stimuli (CS). The assignment of colors to CS+ (squares that might be reinforced with shocks) and CS- (squares that were never reinforced) was counterbalanced across participants. During intertrial intervals, a fixation cross was displayed. The OFL phase consisted of 24 CS+ and 24 CS- trials. During the course of the task, 12 CS+ were reinforced with an uncomfortable shock to the ventral part of the upper right forearm of the demonstrator (unconditioned stimulus, US). The CS+/CS- order was pseudo-random with the restriction that any given CS may not be repeated more than twice. The first and last presentation of the CS+ was always reinforced. Two sequences matching these criteria were created and counterbalanced across participants.

In each trial, the CS was presented on the demonstrator's screen for 9 seconds and the intertrial interval was randomized between 10 and 15 seconds. The US, administered on half of the CS+ presentations, started 7.5 seconds after CS onset and consisted of five unipolar electrical pulses delivered with 1 ms duration and a latency of 200 ms (total stimulation duration = 0.8 s). The learning model's discomfort demonstration started straight after the stimulation administration and was likely to co-terminate with the end of the CS+ display. In order to measure the fear-potentiated startle response, the startle probes were presented to the observer during half of CSs (onsets, randomly chosen, at 6.0, 6.5 or 7.0 seconds after CS onset) and a quarter of intertrial intervals (onsets between 2.0 and 4.5 seconds after fixation

cross onset, in order not to interfere with subsequent CS presentations). The acoustic startle probe was a white noise burst (80 dB(A) and 50 ms duration) presented binaurally through headphones (Sennheiser HD-201). The number of trials, their timing and the reinforcement ratio were adjusted to the future functional magnetic resonance imaging (fMRI) experiment's demands and were based on the fMRI suggestions included in the Haaker et al.'s (2017) protocol.

Biopac STM100C and STMISOC stimulator modules, driven by a National Instruments USB-6001 analog output card, were used to produce the pulses. Two Ag/AgCl electrodes placed 3.5 cm apart (measured between centers) and filled with electrode gel were used. Electrode placement at the upper ventral part of the forearm was chosen because in most participants it caused muscle flexion and a resulting hand movement, even at non-painful stimulation intensities. The shock level was adjusted for each demonstrator individually prior to the learning session (see procedure section), so that it was experienced as very uncomfortable but not painful.

The OFL phase was followed by the DE phase, which consisted of 12 CS+, 12 CS-, and no US. Here, timing of CSs, intertrial intervals, stimulus order and startle probe presentations followed the same rules as described above. A scheme presenting the experimental design is shown in Figure 1.

In the OFL phase, a HD-SDI security camera and a HDMI converter connected to a computer screen were used to transmit video (without sound) of one participant to the other. Stimulus presentation was controlled using Presentation v19.0 software (Neurobehavioral Systems, Berkeley, CA, USA).

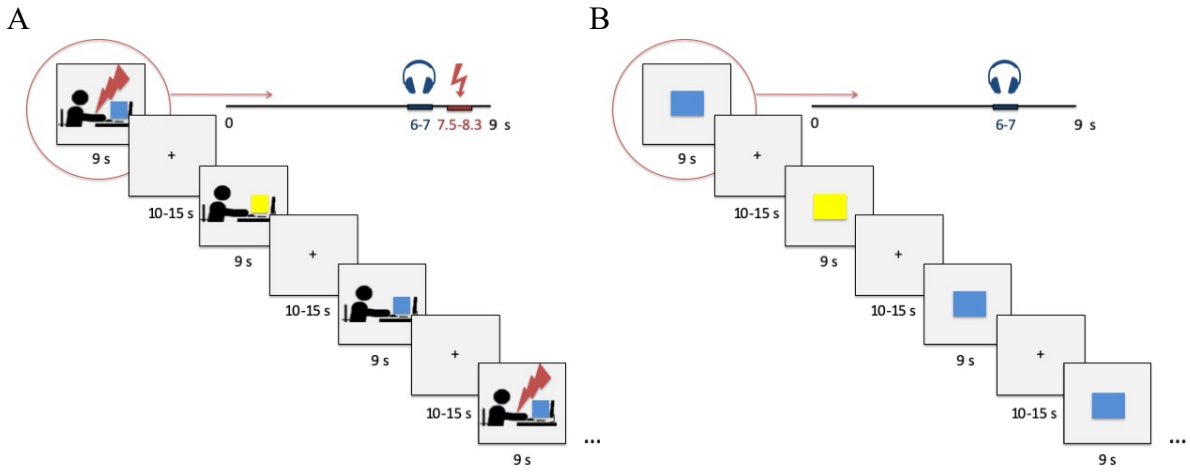

Figure 1. Design of the experiment. (A) In the OFL phase, the observer watched their friend performing a differential fear conditioning task. The conditioned stimuli (blue and yellow squares) were displayed on the demonstrator's screen in a pseudorandom order. Presentation of one of the squares (CS+, here: blue) was accompanied with an uncomfortable electric shock (unconditioned stimulus, US) to the right forearm of the demonstrator with 50% probability. The other color square served as a stimulus that was never reinforced (CS-, here: yellow). A fixation cross was presented between the stimuli. The observer's SCR and FPS response were used to assess learning. In order to measure the startle reflex magnitude, the white noise burst was pseudorandomly presented through the headphones in half of all types of trials (in which a blue square, yellow square, or fixation cross was presented). (B) In the DE phase, the observer performed the demonstrator's task but the USs were not applied. Instructions provided to the observers did not suggest that the electrical stimulation would accompany only one CS in the OFL phase nor that it would be omitted in the DE phase.

Procedure

We used the experimental protocol of Haaker et al. (2017) introducing the following modifications: both demonstrators and observers were invited to the laboratory to take part in the experiment, demonstrators were friends of the subjects, demonstration was transmitted via real time video streaming and fear-potentiated startle was included as an additional measure of conditioning. Upon arrival to the laboratory, the participants were shortly informed about the measurements and stimulation used in the experiment, they filled out the safety form to exclude contraindications for the electrical stimulation and signed the informed consent. Subsequently, they were randomly assigned to their roles as demonstrators and observers and invited to take a seat in one of two adjacent rooms. Skin conductance, electromyography (EMG, used for startle response measurement) and stimulation electrodes were attached and headphones were given to both participants. The demonstrator was seated slightly angled in relation to the computer screen, so that his face, stimulated hand and the computer screen were all visible to the camera relaying the image to the observer's room. Next, the following instruction was given to the demonstrator: *You are the demonstrator. It means that you will be asked to do the task and your performance will be observed by your friend. In the task you will be shown colored figures but only one of them – the blue/yellow one* (depending on the experiment version) – *will be paired with an electric shock administered to the forearm in 50 percent of appearances. It will be very unpleasant but not painful. Please signal the shock occurrences clearly, so that your friend knows that they cause your discomfort. Besides the natural twitch of the forearm, please also try to react with a facial grimace of discomfort, e.g. squint your eyes and frown your forehead. It is very important that you react distinctly only in response to the shock administration – please try not to react to any other stimuli.* Additionally, in order to provide comparable expressions of discomfort across participants, the demonstrator was shown a pre-recorded video of a person

reacting to the shock administration. After the instructions, shock adjustment was performed. Stimulus intensity was gradually increased and after each trial the participant was asked to describe it using an 8-point scale, ranging from imperceptible (1) to painful (8). The adjustment was stopped when the participant described the shock as 'very unpleasant, but not painful' (6). At the time of the demonstrator's preparations, the observer was seated in a separate room. He was asked to sit calmly for 7 minutes while looking at a fixation cross shown on the screen and ignore the sounds presented through the headphones. Fear-potentiated startle and skin conductance signals were recorded during this period, which served as habituation and resting state (baseline) measures. Subsequently, the observer was informed about his role: *You are the observer. It means that you will be asked to watch your friend performing the task. During the task both of you will see colored figures presented on his computer screen and hear short, loud sounds. The sounds will not be associated with the task though, so please try to ignore them. Your friend will receive electric shocks several times during the task. The shocks will be very unpleasant but not painful and they will induce certain reaction in your friend. Please focus on your friend performing the task. Later on, you will be asked to do the same task yourself.*

The OFL phase started with a repeat of the startle habituation (due to interference associated with instructions giving). When the OFL task was over, the video stream was turned off. The observer was then informed that it was his turn to do the same task and the DE phase followed. Identical stimulus material and parameters were used, however, no shocks were administered to the subjects. Finally, the observer filled out an online contingency questionnaire, with questions progressing from open-ended, through percentage ratings of the shock occurrence, to forced choice (G. Weidemann, personal communication, 2 November 2017; Weidemann et al., 2016). Participants were classified as contingency-aware if their forced choice answer was correct and consistent with previous responses. In doubtful

cases, the classification was guided by considering the indicated percentage of trials of a given type associated with shock. These data were used to examine the role of contingency awareness in observational fear conditioning, as this issue has been extensively studied in direct conditioning research (e.g. Grillon, 2002; Hamm & Vaitl, 1996). Additionally, the observer was asked to rate the demonstrator's performance in terms of the level of discomfort expressed as well as the strength and naturalness of reactions; also their level of empathy felt during the observation was assessed. Likert scales ranging from 0 (not at all; very poor) to 9 (very much; very strong) were used for these purposes. In the end, both participants were debriefed about the study.

Physiological recordings

Electrophysiological measurements were collected only from the observers. A Biopac MP-160 acquisition system and AcqKnowledge 4.0 software (Biopac Systems Inc., Goleta, CA, USA) were used to collect the data. Skin conductance was recorded using a Biopac EDA100C amplifier (gain: 5µS/V, 10 Hz low-pass hardware filter) and sampled at 2 kHz. Two 6 mm Ag-AgCl electrodes were attached to the distal phalanges of the second and third digits of the participant's left hand. Startle response was recorded using a Biopac EMG100C amplifier (gain: 2000, 1 Hz high-pass, 500 Hz low-pass hardware filter) and sampled at 2 kHz. Two Ag-AgCl shielded electrodes were attached under participant's right eye, on the orbicularis oculi muscle (one electrode in line with the pupil in forward gaze, the second 1-2 cm laterally). The ground electrode (identical, except non-shielded) was placed on the participant's forehead, directly under the hairline.

Data Scoring and Analysis

Skin Conductance

The recorded signal was decomposed into tonic and phasic components using cvxEDA (Greco et al., 2016). Skin Conductance Responses were scored as the difference

between the maximum value occurring between 0 and 6 seconds after stimulus onset and the mean value from the preceding 2 seconds. The response window was limited to 6 seconds in order to avoid entanglement of the CS and acoustic startle probe effects. While the phasic component should, by definition, have a zero baseline, the subtraction was done to avoid scoring spontaneous fluctuations occurring before the stimulus. Amplitude smaller than 0.2 μS was treated as no response. Due to timing proximity, responses to the US were measured only for trials containing no startle probes during and after CS presentation, and non-reinforced trials (no US) were also scored, by using an identical time window as when the stimulus was present.

SCR data from subjects for whom less than 5 non-zero responses were observed during the DE phase were excluded from the analysis (leading to exclusion of the SCR data of 3 subjects). The resulting amplitudes were normalized within subjects using a log-transformation: $\log(1 + SCR / SCR_{max})$, where SCR_{max} was the highest response found for a given subject. Mean magnitudes (including no-responses as zero amplitude) were calculated for each condition and each participant. Within-subject averages were submitted to group-level analysis.

Fear-Potentiated Startle

Following the guidelines of Blumenthal et al. (2005), the signal was band-pass filtered in the 28 - 500 Hz band (4th order Butterworth filter), rectified (i.e. replaced with its absolute value), and smoothed using a 40 Hz low-pass FIR filter. Each response was scored as the difference between the maximum value in a 20 - 120 ms time window and mean value in the -100 - 0 ms time window (times relative to startle probe onset; trial was scored as 0 if peak amplitude was lower than baseline). Resulting magnitudes were normalized within participants using T-scores. Mean magnitudes were calculated for each participant and each condition and used for group-level analysis.

All trials were visually inspected for the presence of artifacts, which led to exclusion of data from one participant due to an overall noisy recording; however, no individual trials were discarded. EMG data from two other participants, exhibiting 5 or less non-zero responses throughout the entire experiment, were excluded from the analysis (note that different participants were excluded from the EMG and SCR analyses).

Custom Python scripts utilizing Numpy (https://www.numpy.org/), Scipy (Virtanen et al., 2018), and Bioread (github.com/uwmadison-chm/bioread) libraries were used for signal preprocessing and scoring of both skin conductance and startle EMG.

The main measures of interest were the conditioned responses (differences between reactions to CS+ and CS-, both skin conductance and startle) examined in the direct expression phase. PASW Statistics 18 (SPSS Inc., Chicago, IL) was used for statistical analyses.

Results

Behavioral results

Based on the criteria described above, participants' contingency knowledge was assessed. A total of 14 out of 35 observers were classified as contingency-aware and 21 as contingency-unaware. The non-parametric Kruskal-Wallis test ($N = 35$) revealed no differences in the observers' perception of the demonstrators between contingency-aware ($n = 14$) and -unaware ($n = 21$) groups regarding the 4 studied categories: discomfort expressed, strength of reaction, naturalness of reaction, and observer's empathy (see Table 1).

In the group of subjects eligible for SCR analysis, 14 of 32 observers were contingency-aware, while in the group selected for FPS analysis, 13 of 32 observers were contingency-aware.

Table 1

Observer's post-experiment rating of the demonstrator's expression as well as his self-empathy (felt during the observation phase) assessment. Me: median, IQR: interquartile range.

Assessment of	Question	Min	Max	Me	IQR
demonstrator	discomfort expressed	2	8	6	2
	strength of reaction	2	8	6	3
	naturalness of reaction	2	9	8	2
self	empathy felt	0	9	4	4

Skin conductance

In a series of t-test comparisons during the whole group analysis ($N = 32$), we observed a robust increase in observers' skin conductance following the application of electric shocks to their friends. There was a significant difference between reactions measured when observers watched a friend receiving shocks and those recorded during observation of CS+ trials in which no shocks were applied, $t(31) = 8.37$, $p < .001$, $d = 1.69$ (see Figure 2A). No differential reactions to CS+ compared to CS- presentation were found, either in the OFL or in the DE phase.

Having obtained a low ratio of the CS+/US contingency, we decided to check whether the contingency knowledge moderates the relationship between SCRs and the conditioned stimuli (CS+ and CS-). For this purpose, we used a mixed model ANOVA with the skin conductance response as a dependent variable, stimulus type as a within-subject factor, and contingency knowledge as a between-subject factor. In the direct expression phase we found the stimulus (CS+ vs. CS-) x contingency knowledge (known vs. unknown) interaction, $F(1,$

30) = 12.29, p = .001, η_p^2 =.29. Post hoc comparisons (with Bonferroni correction) revealed a stronger skin conductance response to CS+ (M = 0.11, SD = 0.05) as compared to CS- (M = .07, SD = 0.04, p = .002, d = 0.88, 95% CI [0.02, 0.07]) in participants who indicated the CS+/US contingency correctly. Moreover, skin conductance response to CS+ was stronger in the group of contingency-aware (M = 0.11, SD = 0.05) compared to -unaware participants (M = 0.07, SD = 0.05, p = .015, d = 0.80, 95% CI [0.01, 0.08]; see Figure 2B).

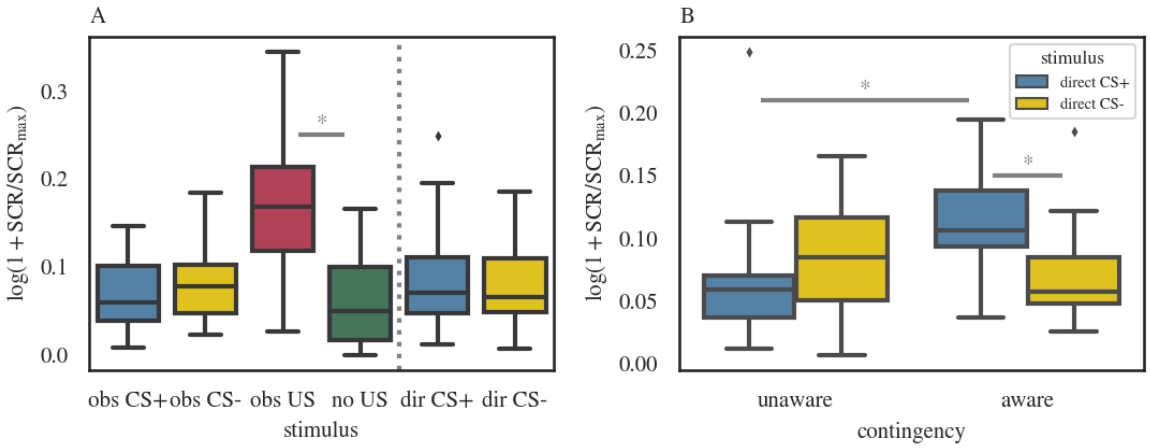

Figure 2. (A) SCR magnitudes to stimuli presented in the experiment: obs CS+ and obs CS- (appearance of CS+/CS- in the OFL phase), obs US and no US (observation of a friend receiving/not receiving electric shock during CS+ in the OFL phase), dir CS+ and dir CS- (appearance of CS+/CS- in the DE phase). N = 32. (B) SCR magnitudes to the appearance of the CS+ and CS- in the DE phase of the experiment for contingency-aware (n = 14) and -unaware (n = 18) participants separately. The stimulus (CS+ vs. CS-) x contingency (aware vs. unaware) interaction was found, F(1, 30) = 12.29, p = .001, η_p^2 = .29, and significant effects of the post-hoc comparisons are marked. Error bars indicate 1.5*IQR beyond the 1st quartile and above the 3rd quartile.

Fear-potentiated startle

In the whole group analysis ($N = 32$), in order to check for the differences in FPS responses to different stimuli depending on the phase of the experiment, we ran a mixed model ANOVA with the startle response as a dependent variable and stimulus type and task phase as within-subject factors. We found the main effect of phase, $F(1, 31) = 81.87$, $p < .001$, $\eta_p^2 = .73$. Post hoc comparisons (with Bonferroni correction) revealed a stronger FPS response in the DE ($M = 54.14$, $SD = 2.59$) compared to the OFL phase ($M = 47.93$, $SD = 1.30$, $p < .001$, $d = 3.03$, 95% CI [4.81, 7.62]; see Figure 3A).

To check whether there was an effect of contingency knowledge on FPS responses to different stimuli in the DE phase, we performed a mixed model ANOVA analogous to the analysis of the SCR data. We found a significant main effect of stimulus (CS+ vs. CS- vs. fixations cross), $F(2, 60) = 8.16$, $p = .001$, $\eta_p^2 = .21$. Further post hoc comparisons (with Bonferroni correction) showed that the FPS reaction to both CS+ ($M = 55.71$, $SD = 4.08$) and CS- ($M = 54.99$, $SD = 5.21$) was stronger than the one measured during the fixation cross presentation ($M = 51.73$, $SD = 4.55$); for CS+ > fixation cross: $p < .001$, $d = 0.92$, 95% CI [1.97, 6.99]; for CS- > fixation cross: $p = .034$, $d = 0.67$, 95% CI [0.22, 6.85]; see Figure 3A. There was also a trend toward the stimulus (CS+ vs. CS- vs. fixation cross) x contingency knowledge (aware vs. unaware) interaction, $F(2, 60) = 2.59$, $p = .083$, $\eta_p^2 = .08$. Results of the post hoc comparisons (with Bonferroni correction) revealed that in the group of contingency-aware participants, the response to both CS+ ($M = 57.13$, $SD = 4.56$) and CS- ($M = 55.01$, $SD = 6.17$) was stronger than the response to a fixation cross ($M = 49.10$, $SD = 3.85$); for CS+ > fixation cross: $p < .001$, $d = 1.69$, 95% CI [3.27, 11.00]; for CS- > fixation cross: $p = .056$, $d = 0.97$, 95% CI [-0.10, 10.12]; see Figure 3B. This trend suggests that the effect of the potentiated startle response to both conditioned stimuli might be specific to only the contingency-aware participants.

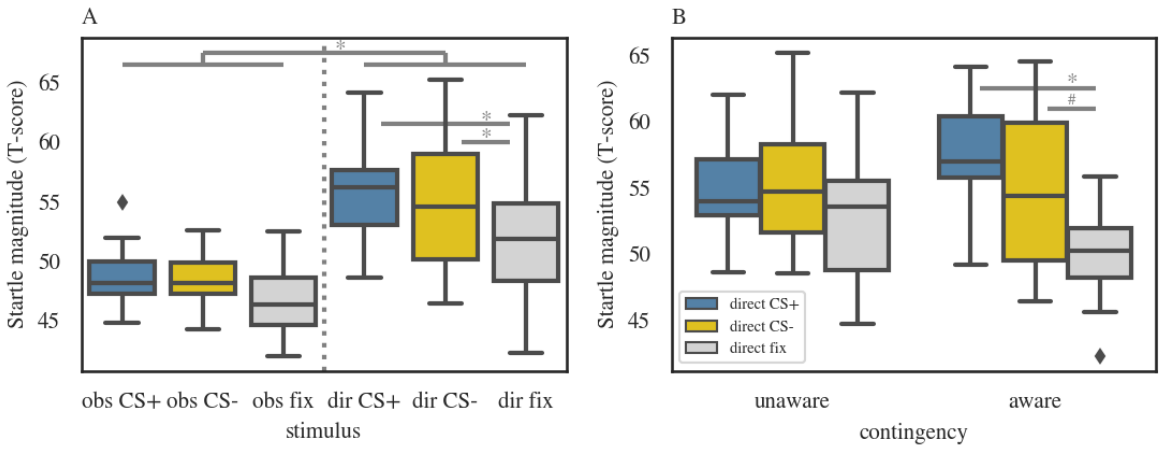

Figure 3. (A) FPS responses measured during different stimuli presented in the experiment: obs CS+, obs CS- and obs fix (presentation of CS+/CS-/fixation cross in the OFL phase), dir CS+, dir CS- and dir fix (presentation of CS+/CS-/fixation cross in the DE phase). $N = 32$. (B) FPS magnitudes measured in the DE phase: results of contingency-aware ($n = 13$) and -unaware ($n = 19$) participants are grouped separately. A trend toward the stimulus (CS+ vs. CS- vs. fixation cross) x contingency (aware vs. unaware) interaction was found, $F(2, 60) = 2.59$, $p = .083$, $\eta_p^2 = .08$, and significant effects of the post-hoc comparisons are marked. Error bars indicate 1.5*IQR beyond the 1st quartile and above the 3rd quartile.

Discussion

In the present study, we used a modified version of the observational fear conditioning paradigm (Haaker et al., 2017) to examine whether the threat learning outcomes can be replicated in more ecological conditions. Most previous studies on observational fear conditioning have used carefully prepared video recordings with actors unknown to the observers. Instead, we arranged a real-time experiment involving participants who were significant to each other. We additionally combined two commonly applied psychophysiological methods, SCR and FPS, in the same paradigm.

We were particularly interested in the results obtained during the direct expression (DE) phase, as they reflected the observational fear learning efficiency. We observed that participants' physiological reactions in the DE stage differentiated between the conditioned stimuli, but the differentiation was dependent on their declarative knowledge of the CS+/US contingency. Specifically, in the case of SCRs, the reactions to directly presented CSs were significantly different only in contingency-aware participants. All participants, however, reacted strongly to the observational US. In the case of FPS, we observed a significant potentiation towards both CSs, but no differentiation between the two. This result suggests the negative affect generalization: it is possible that during the OFL phase participants learned about the aversive value of squares in general, as opposed to the 'safe' fixation cross. As a consequence, during the DE phase, both CSs elicited augmented startle responses. Additionally, we observed a trend-level interaction between contingency and stimulus type, suggesting that the effect of FPS potentiation toward CSs was mostly present in the contingency-aware group. On the declarative level, the general learning efficiency was low, with 14 out of 35 participants correctly identifying the relationship between CS and US.

The physiological results replicate the findings of previous works which manipulated various elements in a video-based vicarious learning procedure and consistently showed differential conditioning effects measured by SCR (e. g. Golkar et al., 2015; Golkar & Olsson, 2017; Olsson et al., 2016) and FPS (Selbing & Olsson, 2019). Interestingly, in the aforementioned studies, an overwhelming majority of the participants correctly identified the CS+/US contingency and the few who did not were excluded from analyses (Golkar & Olsson, 2016). Therefore, the largest difference observed in the current results lies in the low ratio of the CS+/US contingency observed.

The changes we introduced to the protocol of Haaker et al. (2017) were designed to increase its ecological validity. To improve the immediacy and realism of the threat learning

situation, we invited both demonstrator and observer (who were well known to each other) to the laboratory and carried out the observational stage via live streaming. At the same time, the increase of realism came at a cost of lessening our control over experimental conditions: although the demonstrators received general instructions about how they should react to the unpleasant stimulus, each of them still reacted in their own individual way. Such reactions could not be influenced once the experiment started and were not selected through video editing before the experiment (as suggested by Haaker et al. (2017)). While this increased realism, it may also have increased ambiguity, since spontaneous displays of emotions are smaller in amplitude and have different internal timing than posed emotional expressions (McIntosh, 2006). The ambiguity, however, should have been reduced by the fact that the observers were familiar with the demonstrators and thus more likely to be sensitive to their emotional expressions. While our ecologically-aimed modifications likely contributed to the low contingency knowledge, other factors may come into play as well. First, the inclusion of startle probes (cf. Sjouwerman et al., 2016) and low (50%) CS+ reinforcement ratio might have further contributed to the creation of a weak learning situation. It is possible that auditory stimuli not only disrupted participants' attention but also introduced an additional aversive component which interfered with the clear CS+/US association. Increase of the CS+ reinforcement ratio should be considered as a possibility for the contingency learning enhancement. Furthermore, the demonstrators were seated in an ordinary room, rather than against a clear and contrasting background, which might have introduced additional visual distractors.

We find the low level of contingency knowledge and its impact on conditioning measures interesting (as it highlights the role of the observer's attention and the observational stimulus quality), but not surprising, given that the role of contingency knowledge has been described for various conditioning protocols and conditioning measures (e.g. Baeuchl et al.,

2019; Tabbert et al., 2006, 2011; Weidemann et al., 2016). While skin conductance response has been reported as a measure reflecting contingency learning (Sevenster et al., 2014), a potentiation of the acoustic startle reflex has been claimed to be a specific index of fear acquisition, not requiring contingency awareness (Hamm & Vaitl, 1996; Hamm & Weike, 2005) and generally considered to be more valence-specific and less dependent on attention (Lang et al., 1990; although see Mertens & De Houwer, 2016). This study was the first to involve both SCR and FPS measures. We showed that under ecological conditions, observers display general negative arousal to conditioned stimuli but declarative knowledge about the CS+/US contingency is not always acquired.

When taking a closer look at the SCR results, two effects are apparent. First, observational US elicited stronger and more robust reactions than any of the conditioned stimuli in the DE phase. This means that the US was perceived the way we expected – which is important, considering that observing the aversive qualities of the US are a prerequisite for threat learning. Second, differential conditioning of the SCRs was observed only in the contingency-aware participants (explaining the apparent lack of effects when the whole group is considered). This observation could be made because the low rate of learning allowed splitting the participants into two groups and suggests that learning about CS-US contingency can be separated from the response to the observational US.

In the case of FPS results, we observe several effects. Overall, the reactions were stronger in the DE compared to the OFL stage, suggesting that the observers felt an elevated sense of threat in the task in which they expected to experience the aversive stimuli themselves, which confirms a general effect of experimental manipulation. Furthermore, in the DE stage, startle probes presented during both conditioned stimuli elicited heightened responses compared to the ones measured during intertrial intervals, suggesting that the acquired fear was generalized (nonspecific). We observed a trend-level interaction with

contingency knowledge, which upon inspection suggests that the effect is driven by the contingency-aware participants. Although the SCR and FPS results do not show one-to-one correspondence, the information they provide is complementary and consistent.

Finally, it is important to emphasize the significance of this study for further development of the vicarious fear learning line of research. The design proposed here is conceptually similar to the direct threat models of observational fear conditioning in rodents (Jeon et al., 2010; Jeon & Shin, 2011). The main advantage of our design is the real involvement of both demonstrator and observer as well as close relationship between both participants (in rodent studies, subjects are typically cage mates). Furthermore, acoustic startle response is commonly measured in rodent studies and its underlying neural circuit, involving the brainstem and centromedial amygdala, is conserved across species (Koch, 1999; Kuhn et al., 2019). These commonalities give rise to a whole range of possibilities for studying mechanisms of social fear acquisition across species. Research questions that might be answered owing to translational studies involve e.g. comparison of neural circuits related to observational fear learning in humans and rodents, investigation of the impact of participants' familiarity on social fear learning (on both behavioral and neural level), and examination of different channels used for fear transmission (e.g. odor communication in humans).

To conclude, in this work we show that participants watching their friends acquiring fear in real time can learn about the observed threat vicariously although the learning efficiency is lower than reported in previous studies. On the other hand, we found that the negative emotional arousal of all the observers increased following the observational fear learning phase and this emotional reaction was generalized to both conditioned stimuli. The results suggest that the process of learning fear through observation may be composed of two separable components: an automatic, non-specific emotional reaction to the response of the

demonstrator (which can serve as social unconditioned stimulus, US) and learning about predicting stimulus (CS) - US contingency. Further studies are needed to describe the factors underlying successful observational fear learning under ecological conditions.

Data availability

Data from psychophysiological recordings are stored in an OSF repository and are available at https://osf.io/d3wxn/[1]. Code replicating analyses reported here is available at https://github.com/mslw/vic-fear-learning-physio.

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[1] Prior to article publication, project visibility will be set to private. Please use the view-only link: https://osf.io/d3wxn/?view_only=ad87fa2e0076487c92c905f3be933720

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