Running head: AVERSIVE CONDITIONING IN CHILDREN

The use of an unpleasant sound unconditional stimulus in an aversive conditioning procedure with

8 to 11 year old children

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Abstract

The study of aversive Pavlovian conditioning in children can contribute to our understanding of how fears are acquired and extinguished during development. However, methodological issues hamper further research because of ethical and procedural concerns regarding the use of traditional aversive unconditional stimuli (USs) and no established method to measure trial-by-trial changes in the child's expectancy of the US. The present experiment used geometric shape conditional stimuli (CSs) and an unpleasant sound US with 8 to 11 year old children. Reliable acquisition and extinction was observed with first, second, and third interval skin conductance responses, on-line expectancy judgments, and post-conditioning subjective ratings of pleasantness and arousal. The experiment confirms the novel use of an unpleasant sound of metal scraping on slate as a US in aversive conditioning with children. The methods have the potential to facilitate the ethical conduct of aversive conditioning research in children using psychophysiological, affective, and self-report expectancy measures.

Keywords: Pavlovian conditioning; unconditional stimulus; children; extinction; sound

Introduction

Watson and Rayner (1920) showed that fear behaviour to a white rat could be acquired in a child by pairing the rat with an aversive loud noise. In this classic example of aversive Pavlovian conditioning, the white rat is the conditional stimulus (CS) and the loud noise is the unconditional stimulus (US). Subsequent presentations of the white rat alone elicited the conditional response (CR) of fear behaviour in the child. Direct conditioning experiences are a major factor underlying the origin of childhood fears, such as simple phobia and social phobia (Muris, Merckelbach, & Collaris, 1997). Importantly, extinction of fear can be encouraged by repeatedly presenting the feared CS on its own. The clinical application of this process, often termed exposure therapy, plays a central role in behaviour-based treatments of fear and anxiety (Waters & Craske, 2005). For reasons such as these, researchers have sought to understand the mechanisms that underlie aversive Pavlovian conditioning and extinction in children.

When compared to the extensive research conducted in adults, considerably less is known about aversive conditioning in children. Field (2006) presented 9 to 11 year old children with cartoon characters paired with foods that were liked (ice cream) or disliked (Brussels sprouts). After conditioning, the cartoon characters paired with the disliked food were less preferred than those paired with the liked food. Liberman, Lipp, Spence, and March (2006) presented anxious and non-anxious 7 to 14 year olds with cartoon characters paired with a loud tone or presented alone. In the non-anxious children, the character that was paired with the loud tone was rated higher in arousal and fear than the character presented alone after acquisition. Lau et al. (2008) measured fear ratings in anxious and non-anxious adolescents (mean age of 13.64 years) in response to two female faces of neutral expression; one changed to a fearful face paired with a 3 s scream (CS+) and the other remained neutral in expression (CS-). In the control group, the CS+ was rated as more fear-provoking than the CS- after acquisition.

The findings from prior research suggest that children can learn to associate a stimulus with an aversive event. However, there are also limitations and inconsistencies. Prior research has

found aversive conditioning in self-report measures only. Liberman et al. (2006) did not find evidence for conditioning in first interval skin conductance responses. In addition, no experiment has yet measured children's cognitive expectation of the US on a trial-by-trial basis during acquisition and extinction. Finally, inconsistencies across studies have been found when children provide subjective ratings after experimental phases. Field (2006) and Lau et al. (2008) both reported that changes in preferences and fear ratings, respectively, that developed during acquisition persisted across an extinction phase. In contrast, Liberman et al. (2006) found extinction in arousal and fear ratings.

The nature of the US used is a critical methodological consideration for future research in young children. By its definition, an unpleasant US is required to study aversive conditioning (Pine et al., 2001). However, this can present both procedural and ethical problems. Food items such as those used by Field (2006) appear ethically acceptable, but are not in themselves aversive. These stimuli evoke negative associations and it is questionable whether they will evoke similar reactions in all children. The loud tone US used by Liberman et al. (2006) does evoke a direct aversive reaction. However, loud tones may cause pain and distress in children sensitive to loud stimuli (e.g., some anxiety disorders; American Psychiatric Association, 1994). In addition, this stimulus yielded weak conditioning effects in Liberman et al.'s (2006) study. Experiments with adults traditionally use electric shock and generally yield strong conditioning effects (see Grillon, 2002; Lissek et al., 2005). Such stimuli are mildly painful but present procedural and ethical issues when applied to children. Other unpleasant stimuli, such as odours (e.g., Flor, Birbaumer, Hermann, Ziegler, & Patrick, 2002) and air puff (e.g., Suboski, 1967), can be difficult to administer because they need special apparatus and compliance, which may be difficult for children (e.g., refrain from movements).

Neumann and Waters (2006; Neumann, Waters, & Westbury, 2008) showed that the sound of metal scraping on slate, which resembles the sound of fingernails running down a chalkboard, supported the acquisition and extinction of CRs in adult college students and in a sample of

adolescents. The unpleasant sound would appear suitable to study aversive conditioning in children because it is presented at a moderate intensity and it does not require special behaviour from the child (e.g., to remain very still). The present experiment, therefore, aimed to test whether the unpleasant sound can function as a US to support the acquisition and extinction of CRs in a differential aversive conditioning procedure with 8 to 11 year old children. One CS (CS+) was paired with an unpleasant sound during acquisition and a second stimulus (CS-) was presented alone. Both stimuli were presented alone during extinction. Skin conductance responses and on-line ratings of US expectancy were measured on a trial-by-trial basis. Pleasantness and arousal ratings were also taken after each experimental phase. Pleasantness ratings reflect changes in the affective properties of the CS during conditioning and arousal ratings can reflect changes in the affective properties of the CS and changes in the anticipation of the US. The children also rated the unpleasant sound for pleasantness, arousal, and interest.

Method

Participants

The final sample consisted of 8 boys and 8 girls aged between 8 and 11 years ($M = 9.63$ years, $SD = 0.72$) that were recruited from a local primary school and through advertisement. Two additional girls were recruited, but had incomplete data sets after withdrawing their participation and are not considered further. The children had a mean of 4.50 years of education ($SD = 0.73$), were born in Australia and spoke English as their first language. Prior to participation, information about the children's anxiety severity was obtained using the parent version of the Spence Children's Anxiety Scale (SCAS-P; Nauta et al., 2004; Spence, 1998) from which a total score was calculated. The mean SCAS-P total score of 15.00 ($SD = 8.12$) is comparable to the mean reported for non-clinical control samples ($M = 14.2$; Nauta et al., 2004). The parent of each child provided written informed consent and were reimbursed AUS$20 for participation. The experimental protocol was approved by the Institutional Human Research Ethics Committee.

Apparatus

The CSs were pictures of a small white square and a large black square presented against a light grey background. The squares were 10 cm and 30 cm squares, respectively, when presented onto a projection area 1.5 m from the participant via a Panasonic Model PT-L557E LCD projector. The US was a 3 s recording of a three pronged garden fork scraped over slate (see Neumann & Waters, 2006) that was presented through Seinnheiser HD-25 headphones and did not exceed a peak intensity of 83 dB(A). Subjective ratings were obtained with a paper-and-pencil adaptation of the self-assessment manikin (SAM; CSEA-NIMH, 1999). Each page contained an image of a CS and three 9-point rating scales that used anchors based on the SAM. The rating scales were for the dimensions of pleasantness (0 = *very unpleasant*, 8 = *very pleasant*), arousal (0 = *very calm*, 8 = *very arousing*), and interest (0 = *very boring*, 8 = *very interesting*). Adaptations of the graphics from the SAM were placed at the end points of each scale. A similar scale was also used to obtain pleasantness, arousal, and interest ratings to the sound US.

On-line sound expectancy judgments and physiological signals were acquired with a PowerLab (ADInstruments, Sydney) Model 4/20 data acquisition system. Expectancy judgments were made via a dial-and-pointer that could be moved about 270˚, where the extreme left was labelled *certain sound will not occur*, and the extreme right was labelled *certain sound will occur*. The dial could be moved between the extremes to indicate varying degrees of certainty. Children used their preferred hand to provide their ratings. Skin conductance was measured with an ADInstruments Model ML116 GSR Amp and MLT116F electrodes attached to the distal phalanx of the first and second finger of the non-preferred hand. Respiratory artefacts on skin conductance (e.g., sneezes, coughs) were monitored via an ADInstruments Model MLT1132 Piezo Respiratory Belt Transducer. All signals were acquired at 1000 Hz and saved for later analysis using a Dell Optiplex Model GX270 computer.

Procedure

Following an introduction to the laboratory and washing of the child's hands, preparations for the psychophysiological recordings were made. A 3-min rest period followed in which the child was asked to sit quietly while psychophysiological recordings were taken. After the rest period, the children were told that they would see shapes on the screen in front of them and also hear an "unusual sound" through the headphones. The children were further instructed on the use of the US expectancy dial. They were asked to move the dial to indicate whether they expected the "unusual sound" to occur or not. Children were asked to use the dial at all times during the experiment, but particularly whenever a shape appeared. All children were given an opportunity to move the dial around for familiarisation purposes prior to the experiment. The experiment proper was divided into three phases: pre-exposure, acquisition, and extinction. The pre-exposure phase contained two presentations each of the CS+ and CS- and no presentations of the US. After the pre-exposure trials, the experimenter entered the room and obtained subjective ratings for each CS. The acquisition phase was next conducted in which there were 12 presentations each of the CS+ followed by the US and the CS- presented alone. Subjective ratings of the CSs were also taken at the end of this phase. Finally, the extinction phase consisted of 12 presentations each of the CS+ presented alone and the CS- presented alone. At the conclusion of extinction, subjective ratings of the CSs and sound US were taken. The US expectancy judgements and skin conductance responses were recorded during each presentation of the CS+ and CS- throughout all phases of the experiment.

In each phase, each CS presentation lasted 8 s and the nature of which shape served as the CS+ and the CS- was counterbalanced across participants. The onset of the US coincided with CS+ offset in the acquisition phase. The order of the CS+ and CS- presentations were randomised with the restriction that the first CS presented in each phase was counterbalanced across participants. The intertrial intervals varied at random from 13 to 16 s (CS offset to CS onset).

Stimulus presentation was controlled by a Dell Optiplex Model GX270 computer fitted with a

SoundMAX Integrated Digital Audio sound controller.

Data scoring

The expectancy of the sound US was defined as the maximum deflection from the

midpoint of the scale during the CS presentation. The judgment scale had an arbitrary range and

end points. For this reason, expectancy judgments were converted to a scale ranging from -100 to

+100 with a midpoint of zero. Scores of -100 indicate the extreme left of the scale and indicate the

maximum expectation of no sound US and scores of +100 indicate the extreme right of the scale

and indicate the maximum expectation of the sound US. Skin conductance responses were scored

as the distance from trough to apex of the curve that began within a specified latency window

following CS onset. First interval responses (FIRs) used a latency window of $1 - 4$ s, second

interval responses (SIRs) used a latency window of $4 - 9$ s, and third interval responses (TIRs)

used a latency window of $9 - 13$ s. The TIRs reflect the unconditional response to the sound US

for the CS+ during acquisition and omission of the sound US for the CS+ during pre-exposure and

extinction and for all presentations of the CS-. All SCRs were square root transformed to

normalise the distributions. Expectancy judgments and skin conductance responses during each

experimental phase were averaged into two blocks of trials before being analysed with ANOVAs

that employed Greenhouse-Geisser adjusted degrees of freedom for within-subjects factors of

more than two levels. Pair-wise comparisons were used for further investigation of significant

effects. The comparisons used *t*-tests that were adjusted for the accumulation of Type I error by

using Šidák's multiplicative inequality. Analyses employed a two-tailed α-value of .05.

<center>Results</center>

Expectancy judgments

The on-line judgments of the expectancy of the sound US during each experimental phase

are shown in Figure 1. Expectancy did not differ between the CS+ and CS- during pre-exposure, *t*

$(15) = 1.07, p = .30$. However, expectancy of the sound US during the CS+ and expectancy of no

sound US during the CS- developed during acquisition. A 2 x 6 (CS x Block) ANOVA confirmed the development of differential expectancy with a main effect for CS, $F(1, 15) = 45.65$, $p < .0005$, $\eta_p^2 = .75$, and a CS x Block interaction, $F(5, 75) = 15.31$, $p < .0005$, $\varepsilon = .48$, $\eta_p^2 = .51$. Subsequent comparisons showed that the CS+ and the CS- did not differ in Block 1, $t = 2.24$, $p > .05$, but expectancy of the shock was higher during the CS+ than during the CS- in Blocks 2 to 6, all ts > 5.56, $p < .01$.

During extinction, expectation of no sound US developed during the CS+. A 2 x 6 (CS x Block) ANOVA yielded a main effect for Block, $F(5, 75) = 19.19$, $p < .0005$, $\varepsilon = .40$, $\eta_p^2 = .56$, and a CS x Block interaction, $F(5, 75) = 3.03$, $p = .033$, $\varepsilon = .66$, $\eta_p^2 = .17$. Expectancy of the sound US was higher during the CS+ than during the CS- in Block 1, $t = 3.76$, $p < .01$, but not in subsequent blocks, all ts < 1.31, $p > .05$. The difference between the CS+ and CS- in the first block of extinction also appears to be substantially smaller than that in the last block of acquisition. This difference may indicate that there was a generalization decrement across phases. However, it may also reflect that the first extinction trial block contained both Trial 1 and 2 of extinction. A repeated measures t-test was thus conducted to compare the CS+ and CS- on Trial 1 in extinction. A significant difference was obtained, $t(12) = 2.58$, $p = .02$, indicating that there was generalization of acquisition learning across phases.

Insert Figure 1 about here

Skin conductance responses

As shown in Figure 2, robust acquisition and extinction of skin conductance conditioned responses was found. The CSs did not differ during pre-exposure for FIRs, SIRs, or TIRs, all ts < 1.38, $p > .05$. Separate 2 x 6 (CS x Block) ANOVAs were conducted for each skin conductance interval measure during acquisition and extinction. The acquisition of FIRs was confirmed by a main effect for CS, $F(1, 15) = 10.06$, $p = .006$, $\eta_p^2 = .40$. Likewise, a main effect for CS, $F(1, 15)$

= 15.23, p = .001, η_p^2 = .50, confirmed the acquisition of SIRs. As expected, TIRs were larger during the CS+ than during the CS- during acquisition, main effect for CS, F (1, 15) = 29.32, p < .0005, η_p^2 = .66, and TIRs also declined across trials, main effect for Block, F (5, 75) = 2.72, p = .04, ε = .72, η_p^2 = .15.

During the extinction phase, there were no significant main effects or interactions, all Fs < 3.82, p > .05, confirming that extinction of skin conductance conditioned responses occurred. Similar to the US expectancy ratings, repeated measures t-tests were conducted to test whether responses were larger for Trial 1 in extinction. The analyses indicated that FIRs were significantly larger during the CS+ than the CS-, t (12) = 3.04, p = .01, whereas there were no significant differences in SIRs or TIRs, both ts < 1,

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Insert Figure 2 about here

Subjective ratings

The subjective ratings taken at the end of each experimental phase were examined with separate 2 x 3 (CS x Phase) ANOVAs. Figure 3 shows the pleasantness and arousal ratings. The analyses for the pleasantness ratings yielded a main effect for CS, F (1, 15) = 8.77, p = .01, η_p^2 = .37, and a CS x Phase interaction, F (2, 30) = 12.15, p < .0005, ε = .88, η_p^2 = .45. As shown in Figure 3 (top panel), the CS+ was rated as less pleasant than the CS- after the acquisition phase, t = 6.10, p < .01, whereas the CSs did not differ after the pre-exposure or extinction phases, both ts < 0.48, p > .05. A similar pattern emerged with the arousal ratings (Figure 3, bottom panel). The analyses yielded a main effect for CS, F (1, 15) = 14.45, p = .002, η_p^2 = .49, a main effect for Phase, F (2, 30) = 10.19, p = .001, ε = .88, η_p^2 = .41, and a CS x Phase interaction, F (2, 30) = 10.89, p = .001, ε = .88, η_p^2 = .42. The interaction reflected that the CS+ was rated as more arousing than the CS- after acquisition, t = 7.43, p < .01, whereas there was no difference after the pre-exposure or extinction phases, both ts < 2.03, p > .05. The interest ratings to the CSs in pre-

exposure (CS+: $M = 3.38$, $SD = 2.42$; CS-: $M = 3.94$, $SD = 2.46$), acquisition (CS+: $M = 4.13$, $SD = 2.73$; CS-: $M = 2.69$, $SD = 2.18$), and extinction (CS+: $M = 2.69$, $SD = 3.00$; CS-: $M = 2.13$, $SD = 2.63$) showed a pattern in which the CS+ was rated as more interesting than the CS- only after the acquisition phase. However, the CS x Phase interaction failed to reach the preset level of significance, $F (2, 30) = 2.82$, $p = .08$, $\varepsilon = .92$, $\eta_p^2 = .16$.

Insert Figure 3 about here

The subjective ratings to the unpleasant sound that were taken at the end of the experiment showed a mean pleasantness rating of 1.25 ($SD = 1.57$, range = 0 to 6, $CI_{95} = 0.41$ to 2.09). The fact that the 95% CI did not encompass zero indicated that while the mean rating was significantly above zero (i.e., a rating of *very unpleasant*), it was nevertheless rated low on pleasantness. The mean arousal rating was 6.43 ($SD = 1.50$, range = 2 to 8, $CI_{95} = 5.64$ to 7.24) and the mean interest rating was 5.31 ($SD = 2.60$, range = 0 to 8, $CI_{95} = 3.93$ to 6.70) and indicated that the sound was highly arousing and moderately interesting.

Discussion

The unpleasant sound of metal scraping on slate was rated as high in unpleasantness and arousal, thus suggesting that the sound had unpleasant qualitative features in the sample of 8 to 11 year old children. When applied in an aversive conditioning procedure, the sound supported the acquisition and extinction of CRs. Conditioned responding was acquired in an on-line verbal measure of US expectancy, a physiological non-verbal measure (FIRs and SIRs), and subjective ratings of pleasantness and arousal. The effect sizes (η_p^2) obtained for the difference between the CS+ and CS- during acquisition for US expectancy, FIRs, and SIRs, were .75, .40, and .50, respectively. These values are comparable to or even exceed those obtained by Neumann and Waters (2006) in their experiment with adult college students (0.80, 0.33, and 0.27, respectively) and with Neumann et al. (2008) in their experiment with 13 to 17 year old adolescents (0.95 and

0.39 for US expectancy and FIRs, respectively). Taking the results of the three studies together, the unpleasant sound would serve as a reliable US for researchers who wish to study aversive conditioning across all ages of development or in children who have psychological disorders such as anxiety disorders.

A significant decline in TIRs across trial blocks was observed during acquisition. The TIR during acquisition reflects the unconditional response to the unpleasant sound for the CS+ and the effects of the omission of this stimulus for the CS-. The decline in TIRs indicates that the children habituated to the unpleasant sound stimulus. Neumann and Waters (2006) also observed a decline in TIRs across acquisition in a sample of adults. In their study, although a shock US elicited larger overall TIRs than did the unpleasant sound US and a loud tone US, a similar rate of habituation was found for all USs. Taken together, it would appear that participants of all ages habituate to the unpleasant sound stimulus and that habituation of the unconditional response is a property shared across a range of USs.

The extinction of CRs was found in all measures. The extinction in subjective ratings is consistent with our previous results using adolescent participants (Neumann et al., 2008) and with Liberman et al. (2006). The present results differ, however, from Lau et al. (2008) in which no extinction of fear ratings was found to a fearful face CS paired with a scream in an adolescent sample. They also differ from Field (2006) in which resistance to extinction was found in children during an experiment that paired novel cartoon characters to liked or disliked foods. Although it has been argued that the failure to find extinction during aversive conditioning might reflect the process of evaluative conditioning (De Houwer, Thomas, & Baeyens, 2001), methodological differences across the experiments might also be implicated. Further research is needed to test whether features, such as the nature of the US, types of CSs, and number of trials, is important to observing extinction of CRs in children. For instance, the methods of the present experiment could be replicated by using a second group of children in which the US is a disliked food.

The present study also contained novel methodological features. It was the first to employ a trial-by-trial measure of US expectancy in children. Although a common measure with adults (e.g., Neumann, Lipp, & Siddle, 1997; Neumann & Waters, 2006), it was unknown whether children could provide reliable judgements. The smooth pattern in the acquisition and extinction of expectancies (see Figure 1) suggest that this measure has potential future use in research with children. Less distinct graphics were used as the CS+ and CS- (squares of different size and color) than used previously (e.g., cartoon characters; Field, 2006). The advantage of using shapes of different sizes is that a continuous series of intermediate stimuli can be constructed to examine the generalization of conditioned fear in children. It is assumed that conditioned fear will generalize to other similar stimuli (see Watson & Raynor's 1920 early demonstration) and children with anxiety disorders may show an increased tendency to generalize their fear response to other stimuli or situations.

The results of the present experiment and prior research using similar stimuli (Neumann & Waters, 2006; Neumann et al., 2008) or samples with similar ages (Field, 2006; Lau et al., 2008; Liberman et al., 2006) suggest a number of recommendations to study aversive conditioning in children or across the lifespan. The unpleasant sound US is recommended on the basis that it is more ethically acceptable than electric shock and loud tones. It is also less influenced by an individual's prior history than stimuli such as food types or a human scream. We presented the unpleasant sound via headphones. Presentation via speakers should also be effective and would allow aversive conditioning to be studied when headphones are not practical (e.g., in *f*MRI studies). In applying the unpleasant sound, visual CSs are recommended so that different modalities are used to reduce potential confusion in the child and help maintain their attention to the task.

The acquisition and extinction in CRs with the unpleasant sound US has been supported in a range of measures such as skin conductance responses, heart rate, startle eyeblink responses, expectancy judgements, pleasantness ratings, and arousal ratings. Psychophysiological measures

are preferred because they are non-verbal, objective, and less prone to demand characteristics. Requesting children to make US expectancy judgements may also change some aspects of the task by promoting cognitive processing of the CS and US. Subjective ratings taken at the end of experimental phases are simple to employ, but are not sensitive to trial-by-trial changes in learning. Moreover, the need to take ratings may interrupt the progression of the experiment. In the present experiment, CRs early in extinction appeared to show a generalization decrement from the acquisition phase. Although analyses indicated that CRs were larger during the CS+ than the CS- on Trial 1 of extinction for US expectancy and FIRs, the difference was not significant for SIRs. This suggests some generalization decrement in SIRs, possibly due to the interruption caused by asking the children to make subjective ratings after acquisition. Finally, the number of trials used in an experiment will represent a trade-off between ensuring adequate exposure to the stimulus contingencies and reducing the effects of boredom or fatigue. On the basis that differential CRs were evident in all trial-by-trial measures (US expectancy, FIRs, and SIRs) by Trial 8 (i.e., Block 4 in Figures 1 and 2) it is recommend that experiments with children use a minimum of 8 trials and up to 12 trials.

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Author Notes

Figures

Figure 1. Mean expectancy of the unpleasant sound unconditional stimulus to the CS+ and CS-
for each block of trials in the pre-exposure, acquisition, and extinction phase. P = pre-exposure
block, A1 to A6 = the six acquisition blocks, and E1 to E6 = the six extinction blocks. Error bars
depict the standard error of the mean.

Figure 2. Mean first interval (top panel), second interval (middle panel) and third interval (bottom
panel) skin conductance responses during the CS+ and CS- for each block of trials in the pre-
exposure, acquisition, and extinction phase. P = pre-exposure block, A1 to A6 = the six acquisition
blocks, and E1 to E6 = the six extinction blocks. Error bars depict the standard error of the mean.

Figure 3. Mean subjective ratings for pleasantness (top panel) and arousal (bottom panel) after the
pre-exposure, acquisition, and extinction phases. Higher values indicate higher ratings of
pleasantness and arousal. Error bars depict the standard error of the mean.