

Unblocking in Pavlovian Fear Conditioning

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Six experiments used rats to study blocking and unblocking of fear learning. An excitatory stimulus (A) blocked fear learning to a neutral stimulus (B). Unblocking of B occurred if the AB compound signaled an increase in unconditioned stimulus (US) intensity or number. Assessments of associative change during blocking showed that more was learned about B than A. Such assessments during unblocking revealed that more was learned about B than A following an increase in US intensity but not US number. These US manipulations had no differential effects on single-cue learning. The results show that variations in US intensity or number produce unblocking of fear learning, but for each there is a different profile of associative change and a potentially different mechanism.

Keywords: associative change, surprise, prediction error

Many contemporary theories assume that learning during a conditioning trial is a function of predictive error: the discrepancy between the actual outcome of a conditioning trial and the expected outcome of that trial. For example, a rat that is subject to pairings of an auditory conditioned stimulus (CS) with a footshock unconditioned stimulus (US) will only learn to fear that CS if there is a discrepancy between the current associative strength of the CS (V) and the amount of associative strength that the shock US will support (λ). The phenomena of blocking and unblocking have been important in the development of these theories.

Kamin (1968, 1969) subjected rats to CSA–shock pairings. In Stage II, rats received a compound stimulus of CSA and CSB paired with shock. Fear learning to B during Stage II failed despite AB–shock pairings. Blocking of fear learning to B occurred because predictive error during Stage II was low: Subjects could predict the occurrence of shock from A, and so learning about B was impaired. Kamin also demonstrated that blocking could be abolished by an increase in US magnitude from Stage I to Stage II (unblocking). Blocking and unblocking reveal the operation of predictive error, but theorists have differed in their treatment of both the causes and consequences of this error. A common approach has been to treat predictive error as the difference between asymptotic levels of learning supported by the US (λ) and the pooled or summed associative strengths of all CSs present on a conditioning trial (ΣV), or $\lambda - \Sigma V$. The Rescorla–Wagner model (Rescorla & Wagner, 1972) is an example. As a consequence of Stage I training, the associative strength of A is high. Stage II predictive error is commensurately low, and this blocks learning to B. An increase in US magnitude promotes unblocking of B be-

cause it increases Stage II λ and thereby reinstates predictive error. A different approach has been to treat blocking and unblocking as due to variations in the associability of A and B. The Mackintosh model (Mackintosh, 1975) is an example. The model computes predictive error for A ($\lambda - V_A$) and B ($\lambda - V_B$) separately. It suggests that blocking is due to a change in attention paid to the blocked CS (B). Specifically, Stage I training establishes A as a predictor of the US so that at the end of such training, predictive error ($\lambda - V_A$) is small. During Stage II, the attention paid B is initially high and conditioning occurs to it ($\lambda - V_B$). However, on subsequent trials attention to B is reduced because it is a poorer predictor of the US than A. An increase in the Stage II US increases λ and augments predictive error, thereby reducing the difference in predictiveness between B and A so that attention to B is maintained across Stage II and B is learned about.

For the same reasons that these theories make different predictions regarding the locus of blocking and of unblocking, they make different predictions regarding the amount that is learned about the pretrained A versus neutral B during blocking and unblocking. Theories employing common predictive error ($\lambda - \Sigma V$), such as the Rescorla–Wagner model, predict that these two stimuli share associative fate during blocking and unblocking. By contrast, theories employing unique predictive error for each CS ($\lambda - V_A$) and ($\lambda - V_B$), such as the Mackintosh model, allow the associative fate of A and B to differ. There have been no investigations of these different predictions regarding the distribution of learning during unblocking of fear.

The difficulty inherent in such investigations is that differences in the relative amounts of learning about the pretrained A versus the unblocked B during unblocking cannot be inferred from performances to the two CSs presented on their own, due to the fact that such performances, at least for the pretrained A, do not discriminate between Stage I and Stage II learning. This makes it impossible to assess the unique contributions of Stage II learning to the performance observed on test. Rescorla devised a novel procedure to address this problem (e.g., Rescorla, 2000, 2001a, 2001b, 2002a, 2002b, 2003). For example, Rescorla (2001b) trained rats on separate pairings of A and C with a food US in Stage I. In Stage II, rats received pairings of the compound AB

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with food. This is a blocking design. However, rather than test for responding to B, Rescorla tested for responding to a compound of A with the untrained D (AD) and a compound of B with C (BC). The compounds AD and BC, had they been tested at the end of Stage I, would have elicited the same responding because each was composed of a CS that had been reinforced in Stage I (A and C) and a CS that had not (B and D). Any differences between responding to them on test could only be a consequence of what was learned about A and B during Stage II. Rescorla (2001b) reported that responding to BC was greater than to AD. This reveals that more was learned about B than A during Stage II, consistent with the Mackintosh (1975) model. The advantage of this compound test procedure for the experiments described here is that it permits assessment of Stage II learning separately from Stage I learning.

The first aim of these experiments was to use the compound test procedure to study unblocking of fear learning. The second aim was to study the effect of different ways of achieving unblocking (increases in US intensity vs. US number) on learning. In studies of unblocking of fear learning, increases in US magnitude have been produced in two distinct ways. The first has been to increase the intensity or duration of the shock US (e.g., Kamin, 1968; McNally, Pigg, & Weidemann, 2004), whereas the second has been to increase the number of shock USs (e.g., Dickinson, Hall, & Mackintosh, 1976; Dickinson & Mackintosh, 1979; Kamin, 1969; Mackintosh, Bygrave, & Picton, 1977). In both cases, the change in Stage II shock US supports robust learning (i.e., unblocking) to the added CS. However, there has been little direct examination of the consequences of variations in US intensity versus US number for unblocking of fear learning. On the one hand, if the only consequence of an increase in the shock US is to alter asymptotic levels of learning, λ , then provided they are matched on magnitude, there should be no difference between variations in US intensity versus US number. On the other hand, studies of unblocking with reductions in US magnitude during appetitive conditioning have suggested that how such reductions are achieved (change in US number vs. quality) influences unblocking (e.g., Holland, 1984, 1988). The present experiments therefore directly compared how increases in shock US intensity and shock US number influenced unblocking of fear learning.

Experiment 1 studied blocking and unblocking using standard designs. Experiment 2 directly assessed distribution of learning during a blocking design. Experiment 3 assessed the effects of variations in US intensity and US number on single-CS fear learning. Experiments 4a and 4b assessed distribution of learning during an unblocking design with increases in US intensity (Experiment 4a) or US number (Experiment 4b). Finally, Experiment 5 directly compared the consequences of increases in US intensity and US number for the distribution of learning in an unblocking design.

Experiment 1

The aim of Experiment 1 was simply to demonstrate blocking and unblocking of fear learning. The design was a 3×2 factorial. In Stage I, all rats received CSA–shock pairings where the shock US was 0.4 mA in intensity and 1 s in duration. In Stage II, rats received compound conditioning of CSA and CSB (AB) with footshock and also compound conditioning of CSC and CSD (CD)

with footshock. For the block group, the Stage II US was the same as Stage I. For the unblock intensity group, the Stage II US was increased in intensity (0.8 mA, 1 s footshock). For the unblock number group, the Stage II US was increased in number (two 0.4 mA, 1 s footshocks). For the block group, the prior training of CSA should block fear learning to CSB during Stage II. Therefore, less fear should be displayed to CSB than CSD on test (e.g., McNally & Cole, 2006). By contrast, for the unblock groups, the change in Stage II US should prevent such blocking, and therefore equivalent levels of fear should be displayed to CSB and CSD on test (e.g., Cole & McNally, 2007).

Method

Subjects

The subjects were 24 experimentally naïve male Wistar rats. Rats were obtained from a commercial supplier (Gore Hill Research Laboratories, St. Leonards, New South Wales, Australia). They were housed in groups of 8 in plastic boxes (67 cm length \times 40 cm width \times 22 cm height) with food and water continuously available. The boxes were kept in an air-conditioned colony room maintained under natural lighting. Each rat was handled for 3 days prior to the start of the experiment. The experimental procedures followed the ethical guidelines established by the American Psychological Association and were approved by the Animal Care and Ethics Committee of the University of New South Wales.

Apparatus

The apparatus consisted of a set of four identical chambers (24 cm length \times 30 cm width \times 21 cm height). The front and rear walls as well as the hinged lid were constructed of clear Perspex, and the end walls were made of stainless steel. The floor consisted of stainless steel rods, 4 mm in diameter, spaced 15 mm apart (center to center). Each chamber stood 2 cm above a tray of paper pellet bedding (Fibercycle, Mudgeeraba, Australia). The chambers were cleaned with water, and the bedding underneath the chambers was changed between rats. These chambers were located individually within sound-attenuating boxes that were painted white. The boxes were illuminated by a red-light-emitting diode so that levels of illumination within the conditioning chambers were 15 cd/m².

There were two auditory CSs that served as CSs B and D in a fully counterbalanced fashion. An 82-dB (A scale), 750-Hz tone and an 82-dB (A scale), 20-Hz clicker were generated digitally and delivered through speakers mounted in the ceiling of each box. There were two visual CSs that served as CSs A and C in a fully counterbalanced fashion. A constant or flashing (4-Hz) presentation of a white fluorescent light produced an illumination level of 75 cd/m² within the chambers. The light was mounted on the ceiling of each box, immediately above the conditioning chamber. All CSs were 30 s in duration and during conditioning coterminated with the footshock US. This US was a 1-s, 0.4-mA or 0.8-mA unscrambled AC 50-Hz shock from a constant-current generator that was delivered to the floor of each chamber. The current available to each floor could be adjusted with an inline milliampere meter. Digital video cameras were mounted on the rear wall of each box and connected to a digital multiplexer in an adjacent room that, in turn, was connected to a digital video disc

recorder. The stimuli used for conditioning were controlled by computer (LabView, National Instruments, Austin, TX).

Procedure

Preexposure. On Days 1 and 2, all rats were placed in conditioning chambers for 11 min. During this session, rats received four 30-s presentations of each CS at an interstimulus interval of 30 s. Rats were preexposed to the CSs to encourage discrimination between them (Mackintosh & Bennett, 1998).

Stage I. Stage I training occurred on Days 3, 4, and 5, and procedures during Stage I were identical for all groups. Rats were placed in the conditioning chambers for approximately 9 min. During this session, they received two pairings of A with the 1-s, 0.4-mA footshock US. The intertrial interval was random, about a mean of 240 s (min = 191 s, max = 332 s). Three to six hours after each Stage I session, rats were placed in the conditioning chambers for 10 min and no stimuli were delivered. These exposures were intended to reduce the levels of background or context fear.

Stage II. Stage II training occurred on Days 6 and 7. Rats were placed in conditioning chambers for approximately 20 min. During this session, the block group received two pairings of each compound AB and CD with the same footshock used in Stage I. The unblock intensity group received two pairings of each AB and CD compound with a 1-s, 0.8-mA footshock. The unblock number group received two pairings of each AB and CD compound with two 0.4-mA footshocks at a US-US interval of 1 s. The intertrial interval was random, about a mean of 240 s. In addition, rats were placed in the chambers each day 3 to 6 hr after each Stage II session for 10 min, and no stimuli were delivered. These exposures were intended to reduce the levels of background or context fear.

Test. On Day 8, all rats were tested for their fear reactions to CSs B and D. These sessions were 11 min in duration, and rats received four presentations of each 30-s CS in the following order: B, D, D, B, D, B, B, D. These presentations commenced 180 s after placement in the chamber. The interstimulus interval was 30 s.

Scoring and Statistics

Performance during test was recorded. Freezing, defined as the absence of all movement other than that required for respiration (Fanselow, 1980), was scored every 2 s during the 3-min pre-CS period and during the 30 s of each CS presentation. The records of each rat were scored by two observers, one of whom was unaware of each rat's group allocation. The interrater reliability, or the correlation between the percentages of observations scored as freezing for each rat between observers, exceeded .93 in all experiments. The percentage of observations of freezing was then analyzed by means of a planned orthogonal contrast testing procedure. The decision-wise error rate (α) was controlled at the .05 level for each contrast tested using the procedure described by Hays (1972).

Results and Discussion

The mean and standard error of the mean levels of freezing displayed during the test are displayed in Figure 1. (The behavioral design is shown in Table 1.) Mean levels of freezing during the first 3 min of context alone were as follows: block group, 13%

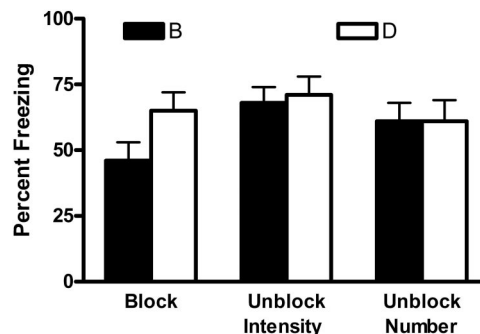


Figure 1. Mean (\pm SEM) levels of freezing on the test for Experiment 1. The behavioral design is shown in Table 1. Fear to B was blocked in the block group but not in the unblock intensity or unblock number group. B and D are two conditioned stimuli (counterbalanced).

($SEM = 6$); unblock intensity group, 9% ($SEM = 4$); unblock number group, 9% ($SEM = 4$). Inspection of the figure indicates that levels of freezing were lower in response to B than D in the block group, but that levels of freezing were equivalent in response to B and D in the unblock intensity and unblock number groups.

The statistical analysis confirmed these observations. Levels of freezing to the CSs were significantly greater than during the first 3 min of context alone, $F(1, 21) = 185.0, p < .05$, and this did not interact with the comparison between the block group versus the unblock intensity and unblock number groups, $F(1, 21) = 3.1, p > .05$, nor did it interact with the comparison between the unblock intensity and unblock number groups, $F < 1$. Overall, there was no significant difference between levels of fear to B and D averaged across groups, $F(1, 21) = 3.9, p > .05$. There was no overall difference in levels of fear between the block group versus the unblock intensity and unblock number groups, $F(1, 21) = 1.1, p > .05$, nor was there an overall difference between the latter two groups, $F < 1$. It is important to note that the difference between fear to B and D interacted significantly with the difference between the block group versus the unblock intensity and unblock number groups, $F(1, 21) = 4.8, p < .05$. This confirms that the blocking of fear to B in the block group was prevented by the change in US intensity or number. There was no interaction, however, between the comparison of performance to B and D, and the difference between the unblock intensity and unblock number groups, $F < 1$. This indicates that the increase in US number and intensity produced equivalent unblocking.

This experiment confirms that prior training of a fear CS can block subsequent fear learning to a novel CS when those stimuli are conditioned in compound. This blocking can be prevented if the introduction of the compound CS coincides with an increase in either the intensity or number of the US.

Experiment 2

Experiment 1 established that fear to a CS can be blocked if that CS is conditioned in compound with a well-trained CS. This experiment used the compound test procedure to assess distribution of learning among the blocked CS (B) and pretrained fear CS (A) during Stage II training. Rescorla (2001b) reported that more is learned about CSB than CSA under similar circumstances in

Table 1
Summary of Experimental Designs

Group	Stage I	Stage II	Test
Experiment 1			
Block	A+	AB+ CD+	B vs. D
Unblock intensity	A+	AB+ CD+	
Unblock number	A+	AB++ CD++	
Experiment 2	A+ C+	AB+	AD vs. BC
Experiment 3	A+ B+ C+ D++		AD vs. BC A, B, C, D
Experiment 4a	A+ C+	AB+	AD vs. BC
Experiment 4b	A+ C+	AB++	AD vs. BC
Experiment 5	A+ C+	AB+ CD++	AD vs. BC A, B, C, D

Note. A, B, C, and D are four conditioned stimuli: a clicker, tone, light, and flashing light, respectively (counterbalanced). + denotes a 0.4-mA shock unconditioned stimulus; ++ denotes two 0.4-mA shocks; + denotes a 0.8-mA unconditioned stimulus.

appetitive conditioning. The aim of this experiment was to extend this finding to fear learning so as to provide a basis for comparison with the unblocking investigated later. In Stage I, A and C were separately paired with footshock, whereas B and D were not trained at this stage. In Stage II, A was presented in compound with B, and the AB compound was paired with footshock. Rats were later tested for their fear responses to the compounds AD and BC. The question of interest was whether learning was distributed equally among A and B during Stage II ($AD = BC$ on test) or unequally ($AD \neq BC$).

Method

Subjects and Apparatus

Sixteen experimentally naïve male Wistar rats were the subjects. They were obtained from the same source and were maintained under the same conditions as the rats in Experiment 1. Rats were handled for 3 days prior to the start of the experiment. All apparatus was as described for Experiment 1. The four CSs used in Experiment 1 (light; 4-Hz flashing light; 82-dB, 750-Hz tone; 82-dB, 10-Hz clicker) were used in this and the remaining experiments. In this and the remaining experiments, rats were tested with a compound of a visual and auditory CS, but the physical identity of each element of the compound was counterbalanced.

Procedure

Preexposure. The procedure was identical to that described for Experiment 1.

Stage I. On Days 3, 4, and 5, rats were placed in the conditioning chambers for approximately 19 min. During this session, they received two pairings of A and C with the 1-s, 0.4-mA footshock US. The intertrial interval was random, about a mean of

240 s. The order in which A and C were presented was counterbalanced within subjects. Three to six hours after each Stage I session, rats were placed in the conditioning chambers for 10 min to reduce fear of the context.

Stage II. On Days 6 and 7, rats were placed in conditioning chambers for 11 min. They received two presentations of the 30-s AB compound followed by the 0.4-mA footshock US separated by a 4-min intertrial interval. Three to six hours later rats were placed in the chambers for 10 min to reduce fear of the context.

Test. On Days 8 and 9, all rats were tested for their fear reactions to nonreinforced presentations of the compounds AD and BC. These sessions were 11 min in duration, and rats were presented with four presentations of each 30-s compound in the following order: AD, BC, BC, AD, BC, AD, AD, BC. These presentations commenced 180 s after placement of the rat in the chamber. The interstimulus interval was 30 s.

Results and Discussion

The mean and standard error of the mean levels of freezing displayed across the course of the experiment are shown in Figure 2. Mean levels of freezing during the 3 min of context alone were as follows: Stage I, 12.3% ($SEM = 3$); Stage II, 11.4% ($SEM = 3.8$); test, 13.4% ($SEM = 3.7$). Inspection of the figure indicates the equivalent acquisition of fear to A and C at the end of Stage I. The figure also suggests that freezing to the compound AB remained relatively stable during Stage II. Finally, it is clear from the figure that rats showed more freezing on both tests to the compound BC than to AD.

Statistical analysis confirmed each of these observations. During each stage, levels of freezing to the CSs were significantly greater than during the first 3 min of context alone, $F_s(1, 15) = 56.6, 104.3, \text{ and } 95.8, p < .05$, for Stage I, Stage II, and test, respectively. At the end of Stage I, there was no difference in levels of freezing to A and C, $F < 1$. There was no change in freezing to the compound AB across days of Stage II, $F < 1$. There was, however, significantly more freezing averaged across the 2 days of test to the compound BC than to the compound AD, $F(1, 15) = 8.3, p < .05$.

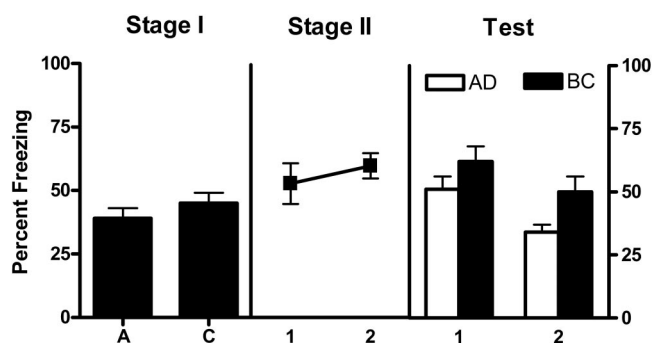


Figure 2. Mean ($\pm SEM$) levels of freezing for Experiment 2. The behavioral design is shown in Table 1. Left: Freezing developed during Stage I to A and C. Middle: Freezing remained relatively stable across Stage II. Right: Performance on the test showed that the neutral B stimulus acquired more associative strength than the excitatory A stimulus during Stage II conditioning (i.e., $BC > AD$). A, B, C, and D are four conditioned stimuli: a clicker, tone, light, and flashing light, respectively (counterbalanced).

These results confirm the findings of Rescorla (2001b). Under circumstances when blocking does occur (see Experiment 1), nevertheless some conditioning does accrue to the blocked CS, and there is actually a greater change to the associative value of the blocked CS (B) than to the pretrained CS (A).

Experiment 3

Experiment 1 showed that fear learning could be unblocked if the introduction of the compound CS coincided with an increase in either the intensity or number of the US. The aim of this experiment was to confirm that those manipulations of the US represented equivalent increases in US magnitude. This was necessary before we could study the distribution of learning during unblocking. To this end, we used the compound test procedure to assess learning to individual CSs conditioned in isolation with USs that varied in either intensity or number.

Subjects were trained with four CSs: A, B, C, and D, which were the same two visual stimuli (light and flashing light) and the two auditory stimuli (tone and clicker) used in Experiment 1, fully counterbalanced. A and C were paired with a 0.4-mA footshock, whereas B was paired with a 0.8-mA footshock and D was paired with two 0.4-mA footshocks. Subjects were then tested for levels of fear to the compounds AD and BC as well as to each element A, B, C, and D. Equal performance to the AD and BC compounds would indicate that the 0.8-mA footshock and two 0.4-mA footshocks supported equivalent amounts of learning, which should also be reflected in performance to the individual stimuli ($B = D$). By contrast, unequal performance to the AD and BC compounds would indicate that the 0.8-mA footshock and two 0.4-mA footshocks supported different amounts of learning, which should also be reflected in performance to the individual stimuli ($B \neq D$).

Method

Subjects

Sixteen experimentally naïve male Wistar rats were the subjects. Rats were obtained from the same source and were maintained under the same conditions as those in Experiment 1. Rats were handled for 3 days prior to the start of the experiment.

Procedure

Preexposure. The procedure for preexposure was as described for Experiment 1.

Conditioning. On Days 3 and 4, rats were placed in conditioning chambers for approximately 35 min. During this session, they received two presentations of each CS. Presentations of A and C were reinforced with the 0.4-mA footshock, presentations of B were reinforced with the 0.8-mA footshock, and presentations of D were reinforced with two 0.4-mA footshocks at a 1-s US-US interval. The order of these presentations was counterbalanced within subjects. Three to six hours after each session, rats were placed in the conditioning chambers for 10 min to reduce fear of context.

Test. On Days 5 and 6, rats were tested for their fear reactions to the compounds BC and AD as well as the elements A, B, C, and D. These sessions were 15 min in duration, and rats were presented with CSs in the following order: AD, BC, BC, AD, A, B, C, D, D,

C, B, A. These presentations commenced 180 s after placement of the rat in the chamber. The interstimulus interval was 30 s.

Results and Discussion

The mean and standard error of the mean levels of freezing displayed during the tests are displayed in Figure 3. Mean levels of freezing during the 3 min of context alone on test was 8.6% ($SEM = 3.1$). From inspection of the figure, levels of fear to compounds AD and BC were equivalent across the 2 days of test. Moreover, fear to the elements B and D were greater than to A and C, although B did not differ from D, and A did not differ from C.

The statistical analysis confirmed these observations. Freezing to the CSs was significantly greater than during the first 3 min of context alone, $F(1, 15) = 220.6$, $p < .05$. There was no overall difference in freezing to AD and BC across the two tests, $F(1, 15) = 2.8$, $p > .05$. Overall, freezing to B and D was greater than freezing to A and C, $F(1, 15) = 73.1$, $p < .05$, although overall freezing to B was not significantly different from freezing to D, $F(1, 15) = 2.5$, $p > .05$, nor was overall freezing to A significantly different from freezing to C, $F(1, 15) = 2.0$, $p > .05$. There was overall significantly less freezing on Test II than Test I, $F(1, 15) = 5.1$, $p < .05$, but none of the contrasts assessing difference in performances to the CSs interacted with this change in freezing, all $F_s < 1$. These results confirm that a 0.8-mA footshock and two 0.4-mA footshocks support equivalent amounts of fear learning to CSs when conditioned in isolation and support significantly greater fear learning than a 0.4-mA footshock. It is unlikely that this equivalence was due to a ceiling effect obscuring potential differences between stimuli. There was significantly less freezing on Test II than Test I, yet the outcome of Test II was the same as that of Test I: Performance to the compounds was equivalent, and performance to the elements revealed $B = D > A = C$.

Experiments 4a and 4b

The aim of these experiments was to study the distribution of learning during an unblocking design involving increases in US number or US intensity. The results of Experiment 1 showed that these manipulations both produced unblocking. However, the design of that experiment did not permit inferences about the distribution of the learning that takes place during unblocking. We have

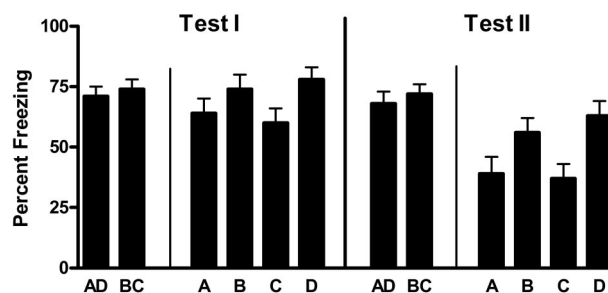


Figure 3. Mean (\pm SEM) levels of freezing on the test for Experiment 3. The behavioral design is shown in Table 1. Freezing to the BC and AD compounds was equivalent, whereas there was more freezing to B and D compared to A and C. A, B, C, and D are four conditioned stimuli: a clicker, tone, light, and flashing light, respectively (counterbalanced).

shown that the USs used to produce such unblocking support nearly identical levels of learning to CSs when conditioned in isolation. The designs of both Experiments 4a and 4b were similar. In Stage I, A and C were separately paired with a 1-s, 0.4-mA footshock, whereas B and D were not trained at this stage. In Stage II, A was presented in compound with B, and the AB compound was paired with footshock. Subjects were later tested for fear responses to the compounds AD and BC. In Experiment 4a, Stage II training employed a 1-s, 0.8-mA footshock, whereas in Experiment 4b, Stage II training employed two 1-s, 0.4-mA footshocks. The question of interest in both experiments concerned the distribution of learning supported by the increased Stage II US. If distributed equally to the pretrained A and unblocked B, then performance to the compounds AD and BC on test should be equivalent. If, however, more is learned about the unblocked B than the pretrained A, performance to the compound BC should be greater than to AD.

Method

Subjects and Apparatus

In both experiments, the subjects were 16 experimentally naïve male Wistar rats. They were obtained from the same source and were maintained under the same conditions as the rats in Experiment 1. Rats were handled for 3 days prior to the start of the experiment. All apparatus was as described in Experiment 2.

Procedure

Preexposure. The procedure for preexposure was as described for Experiment 1.

Stage I. On Days 3, 4, and 5, rats were placed in the conditioning chambers for approximately 19 min. During this session, they received two pairings of A and C with the 1 s, 0.4-mA footshock US. The intertrial interval was random, about a mean of 240 s. The order in which A and C were presented was counterbalanced within subjects. Three to six hours after each Stage I session, rats were placed in the conditioning chambers for 10 min to reduce fear of the context.

Stage II. On Days 6 and 7, rats were placed in conditioning chambers for 11 min. They received two presentations of the 30-s AB compound followed by US separated by a 4-min intertrial interval. In Experiment 4a, the US was 1-s, 0.8-mA footshock. In Experiment 4b, the US was two 1-s, 0.4-mA footshocks with a 1-s US-US interval. Three to six hours later, rats were placed in the chambers for 10 min to reduce fear of the context.

Test. Finally, all rats were tested for their fear reactions to nonreinforced presentations of the compounds AD and BC. These sessions were 11 min in duration, and rats were presented with four presentations of each 30-s compound in the following order: AD, BC, BC, AD, BC, AD, AD, BC. These presentations commenced 180 s after placement of the rat in the chamber. The interstimulus interval was 30 s.

There were three test days in Experiment 4a (Days 8, 9, and 10) and two test days in Experiment 4b (Days 8 and 9). Three test days were used in Experiment 4a because the obtained effect size was small. The third test was not required to achieve statistical significance but instead to indicate robustness of the observed difference.

Results and Discussion

The mean and standard error of the mean levels of freezing displayed on test in Experiment 4a are shown in Figure 4. From inspection of the figure, there was equivalent acquisition of fear to A and C at the end of Stage I. Freezing remained relatively stable across Stage II. Finally, levels of freezing were higher in response to BC than to AD. Mean levels of freezing during the 3 min of context alone were as follows: Stage I, 23.1% ($SEM = 6.3$); Stage II, 28% ($SEM = 5.9$); test, 24.7% ($SEM = 6.4$). During each stage, levels of freezing to the CSs were significantly greater than during the first 3 min of context alone, $F_s(1, 15) = 26.9, 34.1, \text{ and } 34.3, p < .05$, for Stage I, Stage II, and test, respectively. At the end of Stage I, there were no differences in levels of freezing to A and C, $F(1, 15) = 1.2, p > .05$. There was no change in freezing to the compound AB across days of Stage II, $F < 1$. There was, however, significantly more freezing averaged across the 3 days of test to the compound BC than to the compound AD, $F(1, 15) = 10.3, p < .05$.

The mean and standard error of the mean levels of freezing displayed on test in Experiment 4b are shown in Figure 5. Levels of freezing during the 3 min of context alone were as follows: Stage I, 13% ($SEM = 4.8$); Stage II, 11% ($SEM = 4.8$); test, 9.7% ($SEM = 3.2$). During each stage, levels of freezing to the CSs were significantly greater than during the first 3 min of context alone, $F_s(1, 15) = 44.7, 81.8, \text{ and } 123.0, p < .05$, for Stage I, Stage II, and test, respectively. At the end of Stage I, there was no difference in levels of freezing to A and C, $F < 1$. There was no significant change in freezing to the compound AB across days of Stage II, $F(1, 15) = 3.6, p > .05$. Finally, there was no difference in levels of freezing on test to the compounds AD and BC, $F < 1$.

The results of these experiments show that increases in US intensity versus number support different distributions of learning during an unblocking design. An increase in US intensity supported more fear learning to the unblocked CS (B) than the pretrained CS (A). By contrast, an increase in US number supported equal amounts of fear learning to the unblocked B and pretrained A.

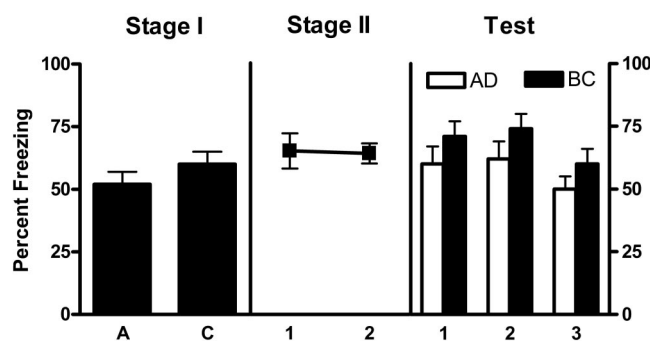


Figure 4. Mean ($\pm SEM$) levels of freezing for Experiment 4a. The behavioral design is shown in Table 1. Left: Freezing developed during Stage I to A and C. Middle: Freezing remained relatively stable across Stage II. Right: Performance on the test showed greater freezing to BC than AD, indicating differences in the amount learned about the unblocked B compared to the pretrained A. A, B, C, and D are four conditioned stimuli: a clicker, tone, light, and flashing light, respectively (counterbalanced).

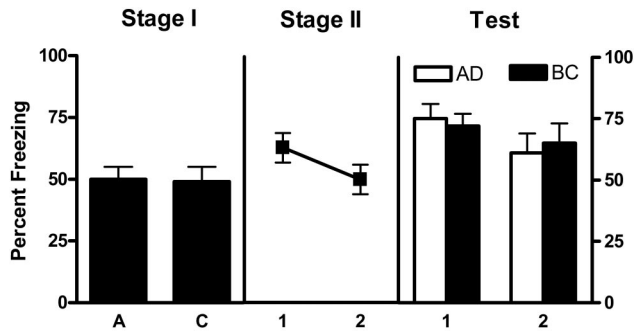


Figure 5. Mean (\pm SEM) levels of freezing for Experiment 4b. The behavioral design is shown in Table 1. Left: Freezing developed during Stage I to A and C. Middle: Freezing remained relatively stable across Stage II. Right: Performance on the test showed equal freezing to BC and AD, indicating no differences in the amount learned about the unblocked B compared to the pretrained A. A, B, C, and D are four conditioned stimuli: a clicker, tone, light, and flashing light, respectively (counterbalanced).

Experiment 5

The results of Experiments 4a and 4b suggested that variations in US intensity and US number can have different consequences for fear learning in unblocking designs. They indicated that an increase in US intensity supports greater fear learning to the unblocked CS than the pretrained CS, whereas an increase in US number supports equal fear learning to both stimuli. One implication of these results, then, is that more is learned about an unblocked CS when it signals a surprising increase in US intensity than when it signals a surprising change in US number. This interpretation relies on the increases in US intensity and number supporting equivalent levels of conditioning (Experiment 3). However, it also relies on comparisons between Experiment 4a and 4b. The designs of those experiments do not permit such comparisons.

The aim of Experiment 5 was to provide a direct comparison between the effects of an increase in US intensity and US number in an unblocking design. Stage I training consisted of A and C presentations paired with a 0.4-mA footshock. In Stage II, the AB compound was paired with a 0.8-mA footshock. Also during Stage II, C was presented in compound with D, and the CD compound was paired with two 0.4-mA footshocks. Rats were then tested for fear reactions to the compounds AD and BC, as well as to the elements A, B, C, and D. The first question of interest was whether the increase in US intensity and number would have similar or different effects on the distribution of learning. Similar effects would be indicated by $AD = BC$, whereas different effects would be indicated by $AD \neq BC$. The second question of interest was whether any such differences were due, at least in part, to differences in the amount learned about the unblocked CS, so that more is learned about the unblocked CS when it signals a change in US intensity. If so, then levels of fear to BC would be greater than AD on test, and levels of freezing to B would be greater than D.

Method

Subjects and Apparatus

Sixteen experimentally naïve male Wistar rats were the subjects. They were obtained from the same source and were maintained

under the same conditions as the rats in Experiment 1. Rats were handled for 3 days prior to the start of the experiment. All apparatus was as described in Experiment 2.

Procedure

Preexposure and Stage I. The procedures for preexposure and Stage I were identical to those for Experiments 4a and 4b.

Stage II. On Days 6 and 7, rats were placed in the conditioning chambers for approximately 20 min. During this session, rats were presented with two pairings of the compound AB with the 1-s, 0.8-mA footshock. They were also presented with two pairings of the compound CD with two 1-s, 0.4-mA footshocks at a US-US interval of 1 s. The order of stimulus presentations was counterbalanced within subjects. The intertrial interval was random, about a mean of 240 s. In addition, rats were placed in the chambers each day during Stage I and Stage II for 10 min without presentation of any stimuli. These context exposures were intended to reduce fear of the context.

Test. On Day 8, rats were tested for their fear reactions to 30-s presentations of the compounds BC and AD as well as to 30-s presentations of the elements A, B, C, and D. These sessions were 15 min in duration, and rats were presented with CSs in the following order: AD, BC, BC, AD, A, B, C, D, D, C, B, A. These presentations commenced 180 s after placement of the rat in the chamber. The interstimulus interval was 30 s.

Results and Discussion

The mean and standard error of the mean levels of freezing displayed across the three stages of the experiment are shown in Figure 6. Mean levels of freezing during the 3 min of context alone were as follows: Stage I, 3.75% ($SEM = 2.5$); Stage II, 15.7% ($SEM = 4.2$); test, 20.8% ($SEM = 6.1$). From inspection of the figure, there was equivalent acquisition of fear to A and C at the end of Stage I. Freezing to AB and CD remained relatively stable across Stage II. On test, rats froze more to the compound BC than to AD. Levels of freezing to A and C, the elements that were trained during Stage I, were equivalent. There was more freezing, however, to B than to D, the elements that were not trained during Stage I. Statistical analysis confirmed each of these observations.

During each stage, levels of freezing to the CSs were significantly greater than during the first 3 min of context alone, $F_s(1, 15) = 68.1, 64.3, \text{ and } 64.7, p < .05$, for Stage I, Stage II, and test, respectively. At the end of Stage I, there was no difference in levels of freezing to A and C, $F < 1$. There was no significant overall change in freezing across days of Stage II, $F(1, 15) = 1.3, p > .05$. There was no overall difference in levels of freezing to AB and CD during Stage II, $F < 1$, and no significant interaction with days, $F(1, 15) = 3.6, p > .05$. Finally, levels of freezing on test were greater to the compound BC than to AD, $F(1, 15) = 12.5, p < .05$. There was no difference in freezing between A and C, $F < 1$, but there was more freezing to B than to D, $F(1, 15) = 6.5, p < .05$. These results show that more fear accrues to an unblocked CS when it signals an increase in US intensity than when it signals an increase in US number.

Taken together, these results suggest that a surprisingly intense shock produces more unblocking than does a surprising second shock. Given these differences in how much was learned about the

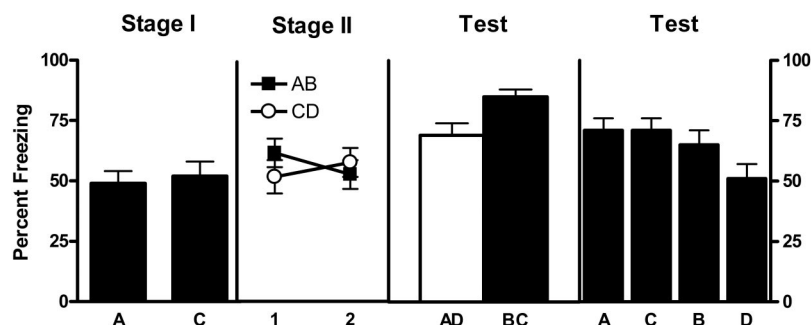


Figure 6. Mean (\pm SEM) levels of freezing for Experiment 5. The behavioral design is shown in Table 1. Left: Freezing developed during Stage I to A and C. Middle: Freezing remained relatively stable to AB and CD across Stage II. Right: Performance on the test showed greater freezing to BC than AD and greater freezing to B than D, indicating that more was learned about the unblocked conditioned stimulus when it signaled an increase in unconditioned stimulus intensity than an increase in unconditioned stimulus number. A, B, C, and D are four conditioned stimuli: a clicker, tone, light, and flashing light, respectively (counterbalanced).

unblocked B and D during Stage II, and the previously documented equivalence of the USs, it might also be expected that less was learned about the pretrained stimulus A than the pretrained C during Stage II. Such differences may well have contributed to the advantage of BC versus AD on compound test. However, there was no evidence for them on the elemental test ($A = C$). This may simply represent the limitation of elemental tests to assess associative change and may underscore the usefulness of the compound test approach. The prior training of A and C in Stage I may have rendered it difficult to detect any differences in Stage II learning using individual presentations of A and C.

General Discussion

These experiments used the compound test procedure described by Rescorla (e.g., Rescorla, 2001b) to study unblocking of fear learning. Experiment 1 showed that prior conditioning of a CS (A) blocked fear learning to a neutral CS (B) when the AB compound was paired with shock. Unblocking of fear learning occurred if the AB compound was paired with a shock larger in intensity or number than that used to condition fear to A in Stage I. Experiment 2 showed, in a blocking design, that more was learned about the blocked CS (B) during Stage II than was learned about the pretrained CS (A). This extends the findings of Rescorla (2001b) to an aversive preparation. Experiment 4a showed, in an unblocking design with increases in US intensity, that more was likewise learned about the unblocked CS B than the pretrained A. By contrast, Experiment 4b showed, in an unblocking design with increases in US number, that there were no differences in the amount learned about the unblocked and the pretrained CSs. Experiment 5 provided a within-subject replication of this difference between learning about increases in US intensity versus US number and suggested that it was due, at least in part, to more being learned about the unblocked CS when it signaled an increase in US intensity. These differences between variations in US intensity and US number were not due to the USs supporting different amounts of learning because Experiment 3 confirmed that the two USs supported equivalent fear learning to neutral CSs conditioned in isolation.

The distribution of learning during the blocking design and the unblocking design with an increase in US intensity are consistent with the Mackintosh (1975) model. Blocking occurs due to withdrawal of attention from the neutral CS (B) during Stage II. This withdrawal takes place on the second and remaining Stage II trials so that learning occurs normally on the first Stage II trial. More is learned about the blocked CS (B) than the pretrained stimulus (A) on this first compound trial because predictive error is greater for B than A. The model predicts a similar distribution of learning during unblocking. It explains unequal distribution of learning among the blocked/unblocked CS and the pretrained CS because those CSs do not share a common associative fate; rather, learning about each CS is determined by its own predictive error, $(\lambda - V_B)$ and $(\lambda - V_A)$. However, for the same reason, the model cannot explain the equal distribution of learning during unblocking with increases in US number.

The finding of equal learning with increases in US number is consistent with the Rescorla–Wagner model. Blocking of fear learning occurs because there is little predictive error during Stage II. The summed associative strengths of A and B are close to the asymptotic levels of learning supported by the US due to the Stage I training of A. In the case of unblocking, the increase in US number supports a greater asymptotic level of learning. This renews predictive error but produces the same associative fate for A and B because the distribution of learning during Stage II is based on common predictive error $(\lambda - \Sigma V)$. This explains the equal distribution of Stage II learning between the pretrained CS and the unblocked CS with an increase in US number, but it cannot explain unequal distribution during the unblocking design with increases in US intensity or during the blocking design.

Le Pelley (2004) and, informally, Rescorla (2001b) have suggested that learning during compound conditioning could be a function of both a common error term, $\lambda - \Sigma V$, as well as a CS-specific error term, $\lambda - V$. For example, during Stage II AB training in a blocking experiment, Le Pelley suggested that more will be learned about the neutral B than the pretrained A because, despite sharing a common error $[\lambda - (V_A + V_B)]$, the CS-specific error term for B, $|\lambda - V_B|$, is larger than for A, $|\lambda - V_A|$. The same approach explains greater learning to the neutral B than the pre-

trained A during unblocking with an increased US intensity. The results reported here are consistent with these suggestions that learning must involve more than just a common error term. Nonetheless, the effects of an increase in shock number promoting equal learning between the pretrained CS and unblocked CS are difficult to reconcile with this approach. One possibility is that attention to the neutral stimulus signaling an increase in US number declines enough to mitigate any effects of the greater CS-specific error term (Mackintosh, 1975), but further experimental work is needed to test this possibility.

A different possibility is that rather than, or in addition to, effects on asymptotic levels of learning, surprising changes in US number themselves directly influence efficacy of the US or CS. Variants of this view have been shared by several authors (e.g., Dickinson et al., 1976; Dickinson & Mackintosh, 1979; Holland & Kenmuir, 2005; Kamin, 1969). For example, in an appetitive arrangement, Holland and Kenmuir reported that the surprising omission of one US enhanced the efficacy of the remaining US. Accordingly, the surprising presence of a second shock may have enhanced the efficacy of the first or both shocks or enhanced the efficacy of the CSs. The present results clearly indicate that alterations of US number can have different effects than alterations of US intensity on unblocking. The difficulty is that more was learned about the unblocked CS when it signaled an increase in US intensity rather than US number—the opposite to that expected if the efficacy of either the US or the unblocked CS had been enhanced by variations in US number.

Finally, these results may be due not just to learning about the CSs as signals for the US, but also to learning about the relationship between the CSs themselves (Rescorla & Durlach, 1981). Stage II of blocking and unblocking experiments allows formation not only of A–US and B–US associations, but also of associations between A and B (Rescorla & Colwill, 1983). In the blocking design, these within-compound associations may benefit B over A because B's associate, A, is a well-trained fear CS, whereas A's associate, B, is less well trained. Such a benefit could produce greater performance to BC than AD. Likewise, in an unblocking design with an increase in US intensity, within-compound associations would again benefit B over A to produce greater performance to BC than AD. The assumption required to explain the present results is that within-event learning is weaker or absent following the increase in US number, rendering the neutral stimulus B unable to benefit. This would produce equal performance to BC and AD. It is important to note that it would establish the differential performance on test to B and D observed in Experiment 5. There is little evidence bearing on this possibility. Studies of second-order conditioning have shown that presentations of a US shortly after compound presentations can disrupt second-order learning (e.g., Cheate & Rudy, 1978; but for examples of facilitation, see Kehoe, Schreurs, & Graham, 1987); and, more generally, posttrial surprise in the form of incongruent training may interfere with association formation (Wagner, Rudy, & Whitlow, 1973). However, the specific assumption required to explain the present results is that within-event learning survives a surprisingly intense shock but not a surprising second shock, and this assumption seems somewhat strained.

In sum, these experiments show that unblocking of fear learning can be achieved via increases in US intensity or US number. These different manipulations of the US have different consequences for

the profile of fear learning. Increases in US intensity favor greater learning to the unblocked CS over the pretrained CS. This distribution of learning is exactly that seen in a blocking design. In contrast, increases in US number support equivalent learning to the unblocked and pretrained CSs. These different profiles of learning can be separately explained by different mechanisms but are difficult to reconcile with the operation of a single associative mechanism during the unblocking of fear learning.

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