Second-order fear conditioning as revealed through augmentation of a startle response: Part I[1]

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A loud, abrupt auditory startle stimulus was presented 1, 17, and 27 sec following termination of a classically conditioned, fear-evoking, second-order stimulus. Forward, second-order conditioned Ss consistently exceeded the startle magnitudes of backward, second-order conditioned controls at all intervals with one exception (the 1-sec interval).

With the possible exception of a study by Brogden (1939), early Russian and U.S. attempts to demonstrate second-order classical conditioning incorporated special reinforcement techniques which made difficult the interpretation of their results (Brogden & Culler, 1935; Eccher & Culler, 1941; Finch & Culler, 1934; Pavlov, 1927). Recently, however, less equivocal demonstrations of second-order conditioning have been reported (Anderson, 1967; Anderson, Plant, Johnson, & Vandever, 1967; Davenport, 1966; Johnson & Anderson, 1969; McAllister & McAllister, 1964). The present study analyzed the possible motivational consequences of a second-order fear response. A technique similar to that of Brown, Kalish, & Farber (1951) was used. They reasoned that any fear-provoking stimulus, as a source of drive, should energize any response tendency that was dominant during or immediately following its presentation. In support of their notion they found that a fear-evoking, first-order conditioned stimulus (CS_1) could augment the magnitude of an unconditioned startle response. The present experiment primarily was concerned with whether the effects of a second-order, fear-producing stimulus (CS_2) paralleled those of a CS_1 as revealed by Brown et al (1951).

SUBJECTS

The Ss were 22 male hooded rats which previously had been employed in another study of second-order learning (Anderson et al, 1967, Experiment 2). They were 135-145 days of age at the beginning of experimentation.

APPARATUS

The chamber in which all first- and second-order conditioning was conducted was constructed of 1/8-in. brass rods which had been molded into circles and mounted flush to the inside surface of three ¼-in. plastic railings. The chamber was 2¾ in. in diam and 6¾ in. long, inside, and was housed inside a larger black enclosure which was located in a ventilated refrigerator shell to insure control of extraneous sources of sound and light. The shock was scrambled, and the CS_1, CS_2 and US contingencies were controlled by silenced programming equipment. The US (135 V ac) was the output of a transformer through a 20 K-ohm, series-connected resistor. The CS_1 was a 6-V buzzer mounted directly in the middle and to the side of the conditioning chamber. The CS_2 was an intermittent 7½-W lamp mounted directly over the center of the conditioning chamber. The CS_2 intermittency (.21 sec on, .21 sec off) was accomplished by additional silenced timing devices.

The test chamber was a stabilimeter-like device which was housed in a wooden electrically shielded box. The chamber was 6½ x 2½ x 3 in. high, constructed of Plexiglas, and mounted on a grid floor (1/8 in. in diam stainless-steel rods, spaced 9/16 in. apart and inserted in plastic railings). The grid floor was suspended at each corner by 1/8-in.-diam flexible 6-in. stainless-steel rods, each of which were connected to respective, firmly anchored, aluminum pillars. Centered and connected to one side of the startle chamber was an Astatic No. 2 ceramic phonocartridge. Inserted in place of the stylus was a 2½-in.-long x 1/8-in.-diam stainless-steel rod, tapered at one end so that it could be fitted into the stylus receptacle. Abrupt, slight displacements of the startle cage generated voltages due to displacements of this stylus. Voltage output was rectified, filtered, and recorded by an ink-writing millivoltmeter (Model 5C Grass polygraph; 5P1 polygraph preamplifier). Preamplifier sensitivity was set at .2 mV/cm for all test sessions except where the magnitude of startle produced pen deflections which exceeded the limit of maximal excursion. The sensitivity then was decreased to either .5 mV/cm or 1 mV/cm. Repeated calibration throughout the experiment indicated that the startle transducer produced an approximate linear output for forces ranging from 2-70 g, dropped from a height of 1 in. from the chamber floor level. Test responses, in general, were within this range.

The startle stimulus was the full output of a Grason-Stadler white-noise generator. Duration was 0.2 sec. The stimuli were presented through a 5-in. Quam speaker mounted directly to the side and 6 in. from the startle chamber.

PROCEDURE

The Ss were maintained in separate cages with free access to food and water throughout the entire experiment. All Ss initially were adapted to the startle chamber for a 10-min period on each of two consecutive days. To obtain a baseline startle response, each S was placed in the chamber for a total of 11 min on each of the next 2 days, the first 5 min of which went uninterrupted. The startle stimulus then was presented five times. The average interstartle interval was 1 min, and magnitude of startle was recorded.

Since approximately 40 days and nine extinction test trials had elapsed since they previously were conditioned, all Ss then were reconditioned. (Prior conditioning had involved 20 first- and 25 second-order trials. A forward-conditioning contingency had been employed on all trials for the experimental group while the controls were treated identically save for a backward contingency for second-order trials only.) In the present experiment a total of 7 first-order trials were semirandomly interspersed with 13 second-order conditioning trials. First-order trials were identical for the two groups, and involved a 5-sec presentation of the buzzer (CS_1) which overlapped and co-terminated with the 2-sec US. For the experimental group (FC_{FC}) second-order trials consisted of a 10-sec presentation of the intermittent light (CS_2) which overlapped and co-terminated with the 5-sec CS_1. For controls (FC_{BC}), the offset of the CS_1 was followed by CS_2 onset 10 sec later. Intertrial interval averaged 60 sec (±15 sec).

The Ss were returned to the startle chamber 24 h later, and, following a 5-min adaptation period, were presented the startle stimulus three times at 1-min intervals to assess possible baseline changes following conditioning. Three additional startle stimuli were presented 1 sec following termination of the 10-sec CS_2. The inter-CS_2 interval averaged 3 min. Three additional test trials were given following 5-min habituation periods on each of the next 2 days. The only differences between the first and next 2 days was that no startle-adaptation trials were given, and the CS_2-startle stimulus interval was 17 sec on Day 2 and 27 sec on Day 3.

RESULTS

The criterion for a response was any pen deflection which exceeded the baseline by 1 mm or more within a ½-sec period from onset of the startle stimulus. Because of the skewed distributions, each response was

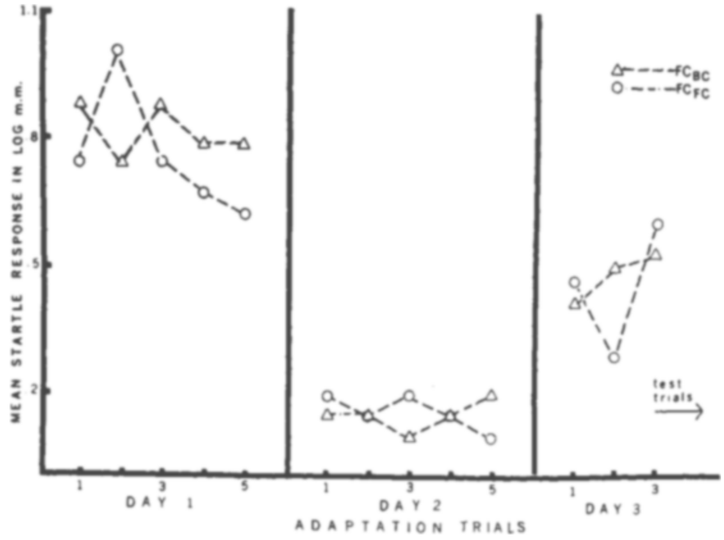

Fig. 1. Mean startle magnitudes (log mm) for experimental (FC_{FC}) and control (FC_{BC}) groups on startle-adaptation trials prior to (Days 1 and 2) and following (Day 3) conditioning.

converted to its logarithm before the data were treated statistically.

Figure 1 summarizes the mean log startle responses for both groups on the startle-habituation trials preceding conditioning (Days 1 and 2) and on the three adaptation trials following conditioning and immediately preceding testing. A three-way ANOVA indicated a near-significant Trials by Days interaction and a reliable days effect ($p < .01$) thus supporting the observation of a decrease in startle magnitude over the three trials of Day 1 but not on Day 2, as well as a pronounced habituation of the startle response for all of Day 2. No other effects were reliable. Marked recovery of startle magnitude for both groups was observed on the three postconditioning, startle-adaptation trials.

Figure 2 summarizes the results obtained on the test trials. With the exception of Test Day 1 the differences in CS_2-produced startle magnitudes were statistically reliable. The forward, second-order conditioned group (FC_{FC}) evinced startle magnitudes which clearly exceeded those of the backward, second-order conditioned controls. A three-way ANOVA produced a significant F for the groups ($F = 7.38$, $df = 1/20$, $p < .025$), days ($F = 13.43$, $df = 1/20$, $p < .01$), and the Groups by Days interaction ($F = 19.10$, $df = 1/20$, $p < .01$) effects. No other effects achieved significance. A grand means analysis (Winer, 1962, p. 56) was performed for Test Days 1 ($F < 1$), 2 ($F = 11.98$, $df = 1/20$, $p < .01$), and 3 ($F = 11.37$, $df = 1/20$, $p < .01$), and indicated that the Groups by Days interaction could be accounted for by the *lack* of differences in startle magnitudes on Test Day 1.

DISCUSSION

These data indicated that a second-order, fear-conditioned stimulus can produce facilitation of a startle response. However, the CS_2-produced startle magnitudes of the forward, second-order conditioned Ss never exceeded those produced by the startle stimulus immediately preceding testing. Apparently, some generalized or residual effect of the reconditioning procedure uniformly altered the preconditioning startle baseline response levels for both groups. It thus would appear that the test performance of the control Ss (FC_{BC}) involved a return to the preconditioning baseline levels, and that the larger startle responses of the experimental Ss on Test Days 2 and 3 represented "true" augmentation rather than perpetuation of the altered baseline which occurred immediately preceding testing. However, this latter interpretation is confounded by the fact that the CS_2-startle stimulus interval was 1 sec on Test Day 1 and 17 and 27 sec, respectively, on Test Days 2 and 3. Thus, conditioning-produced residual activation may have been confounded with the CS_2-startle stimulus interval, as well as with possible extinction of the effect of the CS_2 over days. Moreover, it also could be asserted that the startle-habituation procedure, which preceded testing only on Test Day 1, might have served to "mask" the observed effect had it preceded the other test days. Another experiment thus was felt necessary to evaluate these possible objections.

It may be noted that another objection might be raised because a control for the possible innate aversiveness of the CS_1 was not included. That is, the CS_1 conceivably could have served as a US-like stimulus even in the absence of pairings with shock. However, it previously was found that a control for this possibility was unnecessary for the general procedure we have used (cf. Anderson et al, 1967; Anderson, Plant, & Paden, 1967), that the available data on the aversive properties of buzzers was equivocal (Myers, 1965; Smith, McFarland, & Taylor, 1961), and that the typical result of repeated buzzer presentations was habituation or adaptation. The latter observation probably accounts for the fact that there seems to be no successful reports of classical learning when a buzzer has been the US.

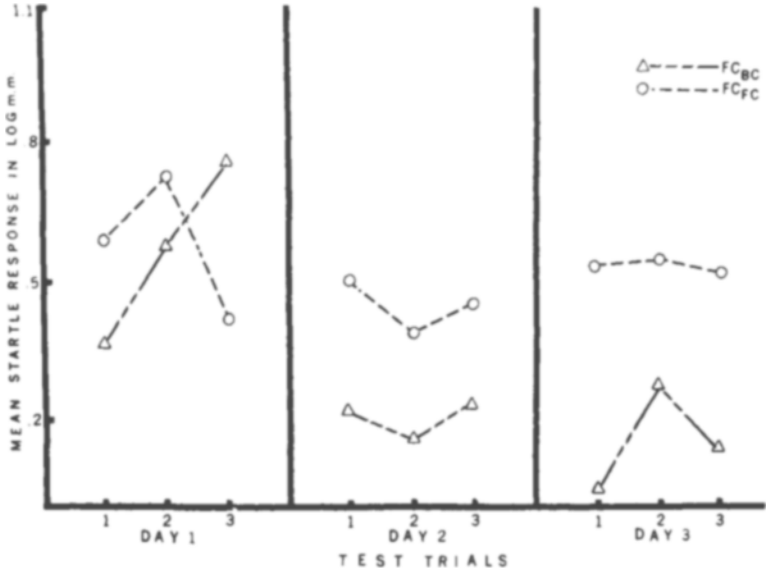

Fig. 2. Mean log startle response magnitudes for each group following offset of the CS_2. The CS_2-startle stimulus interval was 1 sec on Day 1, 17 sec on Day 2, and 27 sec on Day 3.

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NOTES

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Second-order fear conditioning as revealed through augmentation of a startle response: Part II[1]

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An auditory startle stimulus was presented 1, 17, and 27 sec following termination of a second-order, classically conditioned, fear-evoking CS. Forward second-order conditioned Ss consistently exceeded the startle magnitudes of backward second-order conditioned controls at all intervals. An analysis of movement during the CS_2-startle intervals indicated that a postural interpretation could not account for startle augmentation in experimental Ss. A drive-like construct was posited to handle these data.

Anderson, Johnson, and Kempton (1969) found that a classically conditioned second-order stimulus (CS_2) can augment an auditory-produced startle response when the startle stimulus was presented 17 and 27 sec, but not 1 sec, following termination of the CS_2. However, their design confounded both the time of testing and possible extinction of the effect of the CS_2 with manipulation of the CS_2-startle stimulus interval. This study was designed to correct these latter problems.

During the interim between this and the study of Anderson et al (1969), Kurtz & Siegal (1966) provided empirical support for the possibility that a fear-evoking stimulus may augment a startle reaction because of learned postural adjustments (which presumably were conducive to such facilitation), rather than through alterations in motivation level as postulated by Brown, Kalish, & Farber (1951). Kurtz & Siegal (1966) proposed that foot-shock produces postural changes which can become associated with the CS, and that these postural changes are uniquely sympathetic with the startle response. Hence, presentation of the fear-evoking CS in concert with an auditory startle stimulus could produce an augmented startle reaction because of some presumed additive combination of CS-produced postural changes to the startle reaction per se. These authors concluded support for their position because they failed to obtain CS-produced augmentation of startle when the US had been delivered through the back rather than the paws of their Ss during conditioning.

Importantly, these authors neglected the possibility that a back-shock also may have produced unique postural changes, but which could have been antagonistic to the typical startle reaction evinced by the startle stimulus. The CS, as a source of drive, still could have augmented any reaction which had a high probability of occurrence in its presence. Since the back-shock-produced conditioned posture may have been associated with the CS, such would simply have introduced the element of response competition into the startle-augmentation procedure. Interestingly, a more direct test of the postural vs drive-augmentation hypotheses would simply be to tally and analyze the number of movements during the interval between CS termination and presentation of the startle stimulus. No movements during this CS-startle stimulus interval would reflect maintenance of a CS-produced postural adjustment. If movement occurred, such would suggest disruption of the presumed postural adjustment. Further, in order to retain a postural-adjustment hypothesis, the number of movements during the CS-startle interval should be negatively correlated with the magnitude of jump, and that control Ss should move with less vigor. Thus, the present investigation both attempted to unconfound certain features of the study of Anderson et al (1969) and to evaluate the postural vs drive-augmentation hypothesis using the strategy outlined above.

SUBJECTS

The Ss were 32 male albino rats, randomly selected from the experimental and control groups of another study (Johnson & Anderson, 1969). Age and prior experimental experiences at the beginning of this study approximated those of the Ss of Anderson et al (1969).

APPARATUS

The apparatus was that of Anderson et al (1969).

PROCEDURE

The procedure was similar to that of Anderson et al (1969). Adaptation to the stabilimeter chamber involved 3, rather than 2, days of presentation of the startle stimulus in an otherwise identical preconditioning adaptation procedure. Conditioning treatment was exactly the same as for the Ss of Anderson et al (1969). The test procedure was altered in the following manner.

Twenty-four hours following conditioning, the Ss were administered five additional startle stimuli (1-min average interstimulus interval) to determine whether