Effects of Contingent and Noncontingent Shock-Induced Stress Odors

on a Runway Response in Rats

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Abstract

Rats were trained to asymtotic running speed in a straight runway and were then given pretraining with either escapable shock, inescapable shock, or no shock. Subjects were then tested in the runway in the presence of odors from donor rats receiving escapable, inescapable, or no shock. It was suggested on the basis of the results and previous research that inescapable shock stress odor may serve as a conditioned alarm or arousal cue, while escapable shock stress odor may serve as an unconditioned alarm pheromone with highly specific communicative content.

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The mascrosmatic capabilities of the laboratory rat elevates the importance of olfactory cues relative to auditory and visual stimuli in providing information about the environment as was demonstrated by Nigrosh, Slotnick, and Nevil (1975). As the significant influence of odors on rat behavior has become more apparent, the functional properties of odors involved in regulating rat behavior have received increasing research attention (see Shultz & Tapp, 1973, for a review of this area). One avidly studied area is concerned with biological odors generated by rats undergoing stress. Several studies (Dua & Dobson, 1974; King, 1969; Valenta & Rigby, 1968) have demonstrated the capabilities of the rat to discriminate between noncontingent shock-induced stress odors and nonstress odors. Ralls (1971) proposed the existence of an alarm pheromone in the rat, while Stevens and Köster (1971) and King, Pfister, and DiGiusto (1975) made observations indicating that the stress cue was not an alarm pheromone. Sears (1975) using contingent or escapable shock, noncontingent or inescapable shock, and no shock conditions for both background and odors indicated that inescapable stress odor may serve as a conditioned alarm cue and escapable stress odor may serve as an unconditioned alarm cue.

The present study was concerned with further investigating the role of contingent and noncontingent shock-induced stress odors by examining their effect on an appetitive discrete trials runway response. Rats were trained to asymtotic running speed in a straight alley runway and were then preconditioned by administering escapable shock, inescapable shock, or no shock. The rats were then run in the runway in the presence of

odors from donor rats receiving inescapable, escapable, or no shock. All
rats received a constant reward of three 45 mg food pellets on every
trial. It was predicted that the animals would perform differentially as
a function of odorous condition and interaction between odorous condition
and preconditioning.

Method

Subjects

The subjects were 27 male Sprague-Dawley rats approximately 75 days
old at the start of the experiment. All animals were housed individually
with water available ad lib. Subjects were randomly divided into nine
groups of three rats. Three additional rats were used as odorant animals.

Apparatus

The apparatus consisted of a specially constructed double hull
straight alley runway. The alley was constructed of clear plexiglass
with frosted outside surfaces. The outer hull was 203 cm long X 13 cm
high X 46 cm wide. The inner hull was 192.5 cm long X 13 cm high X 10 cm
wide. The inner hull was divided into a 30 cm start section, a 122.5 cm
run section, a 30 cm delay section, and a 10 cm goal section; all of which
were separated by guillotine doors. The inner hull was located in the
center of the outer hull with the start section flush against one end of
the outer hull. The subject's running time in the alley was measured by
three .01 sec Lafayette model 54014 timers, all of which were started by
a microswitch activated by raising the start door. The first timer was
stopped by a photocell 11 cm from the start door; the second timer was
stopped by a second photocell located 34 cm from the start door; the third
timer was stopped by a third photocell located 6.5 cm inside the delay

section. A teaspoon mounted in the middle of the far end of the goalbox served as the foodcup. The apparatus had a plexiglass top which was made secure by means of self-adhesive plastic foam tape and air condition duct tape. Air was exhausted from the runway through a duct at the goal end of the outer hull by means of a fan. Air intake was through a duct at the start end of the inner hull. Located on each side of the inner hull were nine three cm diameter holes which began 50 cm past the start door and ended five cm before the delay door. One cm diameter holes perforated the goal end of the inner hull (13) and the guillotine doors (21). These holes allowed flow of air through the inner hull and into the outer hull. As added olfactory control, paper, which could be replaced after each animal, covered the floor of the inner runway. Background and odorized air were introduced into the inner hull through the top of the runway 30 cm from the start door.

Plexiglass two-way shuttle boxes 35.56 cm X 21.32 cm X 25.40 cm were used for background training (preconditioning) and donor odor collection. The boxes had tilt floors made of stainless steel grids. Microswitches on the floors measured shuttling. The shuttle boxes were housed in Lehigh Valley Electronics sound-insulating chambers. Stimulus parameters, latency measures, and shock presentations were automatically controlled by Lehigh Valley Electronics programming equipment. A Lehigh Valley Electronics Model 113-04 constant current shock source delivered scrambled 1 ma shocks. Odorous air was collected from donor animals below the grid floor of the donor shuttle box and pumped by means of a Universal Electric Company Model 81-012 vacuum pump through 1 cm glass tubing to the runway. An Automatic Switch Company Model 832062 two-way valve between

the vacuum pump and the runway activated by raising the start door and stopped by the third photocell controlled the flow of odorized air into the runway.

The standarized olfactory background air was purified by passage through a dissicant and activated charcoal and provided by a Bell and Gossell air pump model P200 through 1 cm glass tubing to the runway.

Procedure

The subjects were randomly divided into nine groups of three animals each on the basis of the three preconditions and odorous conditions; i.e., naive, inescapable shock, and escapable shock.

All subjects were placed on a food deprivation schedule so that an 85% body weight was established. This percentage was maintained throughout the experiment.

Training phase. All groups received 60 trials (4 trials per day) in the runway with three 45 mg food pellets in the foodcup so that an asymtotic, baseline running speed was established. The runway procedure was identical for all animals in both phases with the exception of odorous condition. The subjects were placed in the startbox facing the door. The start door was raised and the animal was allowed to traverse the runway and enter the goal box (both the delay and goal section doors were raised throughout the experiment). The subject was given time to consume the three pellets and was then removed from the goalbox and placed in a plexiglass holding cage for an intertrial interval of 1 min. After the running of each subject, the paper on the floor of the maze was changed. The olfactory control fan and pumps were on for all runs.

Following the last trial on the last day of training, subjects were

given their preconditioning. The escapable shock subjects (E-R) received 64 unsignalled 5 sec, 1 ma shocks on a VI-30 sec in a shuttle box. Escape was contingent upon shuttling to the other side of the shuttle box. Inescapable shock subjects (I-R) received the same 64 unsignalled 5 sec, 1 ma shocks except no response terminated shock. The naive subjects (N-R) were placed in the shuttle box for 30 min. but were not shocked.

Testing phase. The testing phase began the day following preconditioning and lasted for 10 days (4 trials per day).

The subjects were run in the runway, as in training, but under one of three odor conditions. Odorized air was introduced into the runway from a shuttle box containing one of three donor animals, naive (N-D), inescapable shock (I-D), or escapable shock (E-D). The donor animals had received training as in the three subject conditions for three days prior to the beginning of the testing phase.

As a control procedure, the three donor conditions were run separately from one another. When an animal was removed from the donor or preconditioning shuttle box, air in the chamber was blown by the compartment fan through tubing to a room vent and the shuttle box was washed and dried thoroughly.

Results

Times were computed for the photocell bound sections and were converted into reciprocals. Daily block means of the reciprocals were used as the dependent variable. The results herein reported are for times between the second and third photocells.

An analysis of variance on the last three days of training revealed a significant main effect in running speed of groups formed on the basis

of donor condition (F(2, 18) = 3.79, $\underline{p}<.05$). Posteriori analysis using Scheffe's test indicated that the escapable donor condition groups were running faster than the inescapable donor condition groups ($\underline{p}<.05$) during training. The analysis of variance failed to reach significance for the days' main effect or for any interactions thus indicated that all groups were relatively stable at the end of training.

Change scores (difference from asymtotic baseline reciprocals) were computed for the ten days of testing. The change scores were analyzed using a 3 (donor animal) X 3 (preconditioning) X 10 (daily blocks) repeated-measures analysis of variance (Winer, (1971).

Main effects of donor animal (F(2,18)=7.81, $\underline{p}<.01$) and donor X daily blocks interaction (F(18,162)=2.09, $\underline{p}<.01$) were found to be significant. Although donor X preconditioning (F(4,18)=2.72, $\underline{p}<.07$) only approach significance, experimenter interest led to further post hoc analysis. Using Scheffe's test, comparison of means analysis of donor main effect showed that odor from inescapable shock produced faster running speed than either naive odor ($\underline{p}<.02$) or escapable shock odor ($\underline{p}<.01$). The naive odor condition and escapable shock odor condition were not significantly different in terms of running speed.

Subsequent posteriori analysis of the donor by recipient interaction revealed that when the recipient animals were naive, there was no difference due to odor condition. However, with both the inescapable and escapable background conditions, inescapable shock odor (I-D) produced faster running speeds than either of the other two odor conditions ($\underline{p}s<.01$). Comparisons of recipient animals at a particular level of odor factor revealed that under naive odor conditions (N-D) the naive recipients

ran faster than either the inescapable shock recipient ($p < .05$) or the escapable shock recipient ($p < .01$).

Table 1 contains a summary of the analysis of variance.

Insert Table 1 about here

Discussion

The results of the experiment indicate the discriminability of contingent and non-contingent shock-induced stress odors and their effect on an appetitive runway response. Sears (1975) examined the effects of these odors on escape avoidance learning and the results of the present experiment tend to complement those findings.

The interaction of donor and preconditioning factors supports the hypothesis that odor cues would effect running speed as a function of odorous condition and preconditioning. The increase in running speed for the escapable shock and inescapable shock groups under the inescapable shock odor condition while the naive group did not increase in running speed is consistent with the hypothesis that stress odors from a rat undergoing inescapable shock may serve as a conditionable alarm cue but are not specific, unconditioned alarm pheromone (King et al., 1975; Sears, 1975; Stevens & Köster, 1972). The increased running speed is consistent with the suggestion (Sears, 1975) that the odor serves as a CS eventuating conditioned fear eliciting a species-specific defense reaction (SSDR's) of running (Bolles, 1970). Another possibility is that the odor serves as a conditioned arousal cue which would increase the performance on any task. These hypotheses are testable by examining the effects of the odor

on an operant task. Due to the nature of an operant task, if SSDR's are operating, performance should decrease, whereas if general arousal is taking place, performance should be elevated.

The running speed of all three background groups remained near base-line and there was no significant difference between the groups under the escapable shock odor condition. This may be attributable to the faster running speed of the rats in these groups during training. It is possible that these rats may have been at the upper limit of running speed as suggested by Bower (1961). This "ceiling effect" would prevent any in-creases in running speed that the escapable shock odor might normally cause. Another experiment utilizing shock (passive avoidance) might have affected the baseling running speed of these rats. The experiment was being run during these odor recipient groups training phase. While the passive avoidance experiment was run at a different time of day with attempted olfactory controls, residual odors may have been in the room and been responsible for increasing running speed. In conjunction with Sears' (1975) finding that performance on an escape-avoidance task increased for all three groups under the escapable shock odor condition, this finding may also be due to escapable shock odor condition serving as an uncondition-ed alarm pheromone with very specific communicative content. The speci-ficity suggested could account for the increased performance in the pres-ence of escapable shock odor on an escape-avoidance task in which the odors communicative content would be highly applicable (Sears, 1975). Speci-ficity would also account for no effect of the escapable shock odor on a situation in which the rat is responding for food, such as a runway

response, where escape from shock is not directly appropriate.

The naive background group's faster running speed than either of the shock background groups under naive donor conditions is perhaps due to increased experimental anxiety because of exposure to shock. Both shocked groups are slightly suppressed relative to baseline running speed. The possibility exists that this suppression is due to the generation of higher emotionality by handling the rats and exposing them to an experimental situation after they have been shocked. In Sears' (1975) experiment this hypothesized emotionality due to exposure to shock would not show the same effect because the testing situation also involved exposure to shock.

This study demonstrates the importance of biological odors on situations different from those in which they are emitted. For macrosmates, such as the rat, odor conditions must be a primary concern of control in any type of experiment. Experiments of one nature may have an effect on other experiments taking place in the same location, if proper olfactory controls are not observed. It is necessary to continue investigating the functional significance of odors in order to determine when and how they affect the behavior of the laboratory rat.

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Table 1

3x3x10 Repeated-Measures ANOVA

Source	df	MS	F
A (donor)	2	.9271	7.8127 **
B (recipient)	2	.0709	.5971
AB	4	.3225	2.7177
Error Between	18	.1187	
C (trial Block)	9	.0178	1.4640
AC	18	.0255	2.0934 **
BC	18	.0132	1.0803
ABC	36	.0106	.8723
Error Within	162	.0122	

** $p < .01$