Extinction of the excretory alkali metal response (EAMR) to stress in a reptile[1]

IRWIN M. SPIGEL and ALEX RAMSAY, Erindale College, University of Toronto, Clarkson, Ontario, Canada

Some parameters in the extinction of the stress-induced potentiation of sodium and potassium excretion in turtles were examined in three experiments. A pattern similar to that of a conditioned-response decrement was obtained with termination of shock, and the sensitivity of EAMR extinction to differential antecedent shock administration was demonstrated. It was also found that the increment in cation excretion, following shock administration in a consistent daily pattern, was much more resistant to extinction than that observed in randomly shocked Ss. The difficulty in reconciling these results with those from other species and using other procedures is discussed.

It has been shown in a previous study (Spigel & Ramsay, 1969) that elevated urinary alkali metal levels (combined Na^+ and K^+) followed periodic administration of electric shock to turtles. It has also been demonstrated that this potentiated cation excretion continued for several days in the absence of shock when Ss were retained in the same surround. It was concluded that the sustained higher level of urinary alkali metal was the consequence of a conditioned emotional response to the antecedent shock situation. This finding, however, continues to be at variance with the limited data from rats (Paré, 1964) that indicated that conditioned stress failed to produce significantly elevated cation excretion. Compounding the problem is the positive evidence from human Ss (Hoagland, 1961, pp. 40-63) that related antecedent competitive stress to increased urinary concentrations of sodium and potassium.

The current investigation was designed to examine further the pattern of the observed excretory alkali metal response (EAMR) in the turtle during the course of extinction. Specifically, the experiments sought to determine the relationship of EAMR extinction to differentially applied antecedent shock inasmuch as the original studies utilized only one level of the stimulus condition and a uniform program of administration.

EXPERIMENT 1

Subjects

The Ss were 20 male *chrysemys* approximately 5 years old.

Apparatus

Eight plastic food preserving containers (8 x 4 x 4 in.), with tightly fitted perforated lids, housed the Ss during the experiment. A timer connected to a variable-shock generator was set to deliver a 1.5-V shock of 1-sec duration every 59 sec. The current was administered to Ss by means of insulated alligator clips inserted through holes in the boxes and connected to the tail and right hindleg. The timer was automatically reset by a gear-activated switch. The plastic containers housing the Ss were kept in a wooden compartment (7 x 2 x 1 ft), equipped with two 7.5-W lamps, for the duration of the experiment. An IL flame photometer was employed for Na^+ and K^+ determinations.

Procedure

The Ss were divided randomly into four equal experimental groups, the treatment for each differing with respect to the length of time the shock schedule described for Experiment 1 (Spigel & Ramsay, 1969) was administered. A 24-h light-on condition was retained for the 7 days of the experiment. One group of Ss received 1 sec of shock each minute for 12 h, a second group for 6 h, another group for 3 h, and a fourth group for 1 h. After each of the assigned shock periods, electrodes were removed. Residual habitat water was analyzed for Na^+ and K^+ after each 24-h period and the containers cleaned and refilled as described for Experiment 1 (op cit). In effect, there occurred a 6-day extinction phase for all Ss following differential shock periods on the initial day for the four groups of Ss.

Results

A comparison of extinction curves following differential antecedent shock administration is shown in Fig. 1. Ss given the 12-h shock schedule took longest for excretory alkali metal levels to reach normal—fully 5 subsequent nonshock days. The concentration of excretory cations for the 24-h period initiated by the actual shock administration (Day 1) was directly related to the duration of the antecedent stimulation, the 12-h Ss yielding the highest level and the 1-h group the lowest.

Although the EAMR extinction pattern of 1-h and 3-h shocked Ss did not differ significantly from Days 2 to 7, the 12-h and 6-h groups did excrete significantly higher cation concentrations than the other two groups through the second postshock day (Day 3). The 12-h shocked Ss remained higher in alkali metal output than all others through Day 6.

The analysis of variance for the four groups over the 7 days yielded significant F

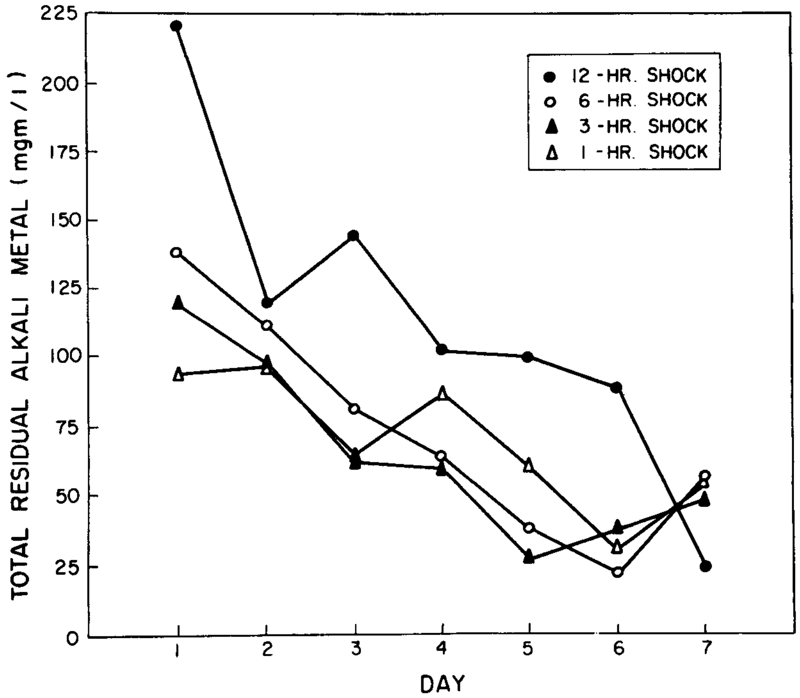

Fig. 1. Comparison of the extinction of the excretory alkali metal response in Ss given varying amounts of electric shock on Day 1.

ratios of 3.58 (df = 3/16), 28.87 (df = 6/96), and 3.25 (df = 18/96), respectively, for groups, days, and the Groups by Days interaction.

EXPERIMENT 2

Subjects

Twelve male *chrysemys*, approximately 5 years old, were the Ss.

Apparatus

Same as in Experiment 1.

Procedure

The Ss were randomly divded into two groups of six. A 24-h light-on cycle was maintained for the 16-day period of the experiment. Because of the length of the experiment, 30 ml of distilled water were added to the containers each day rather than 15 ml as in the two previous experiments. The group designated "random" was administered a total of 48 h of shock experience over a 6-day period, but in the following manner: 4 h on Day 1, 12 h on Day 2, 2 h on Day 3, 8 h on Day 4, 16 h on Day 5, and 6 h on Day 6. That is, Ss were shocked for 1 sec every minute for the specified number of hours each day. Ss designated "nonrandom" also received a 48-h total shock experience over the 6-day treatment period, but for consistent daily periods of 8 h. Following the final shock day, all Ss underwent a 10-day extinction phase. Residual water was analyzed daily for Na^+ and K^+, and the containers refilled with 30 ml of distilled water.

Results

Total excretory alkali metal levels for the randomly and nonrandomly shocked Ss during the 6 days of antecedent stimulation and the 9 subsequent nonshock days are shown in Fig. 2. While there is considerable overlap in the group data during shock days, extinction proceeded much more rapidly for the randomly shocked Ss. It can be seen, in fact, that the excretory cation concentrations of randomly shocked Ss is lower on each of the 9 extinction days, with no overlap whatsoever.

Mann-Whitney U tests of excretory cation levels of the two groups over the shock days (1 to 6) indicated no significant difference. The difference in urinary Na^+ and K^+ output for the two groups for Days 7 to 16, however, was highly significant ($U = 6, p < .001$).

The excretory cation pattern during extinction is further broken down in Table 1, where mean concentrations are shown for three stages of the experiment.

Urinary cation levels for nonrandomly shocked Ss during the 9 nonshock days were not significantly different from those of the 6-day period of antecedent shock administration.

Table 1
Mean Excretory Alkali Metal Levels (mg/1) for Ss During the Six-Day Shock Period and Two Stages of Extinction

Group	Days 1-6	Days 7-11	Days 12-16
Randomly Shocked	100.2	55.2	62.1
Nonrandomly Shocked	90.7	80.8	86.7

DISCUSSION

The results of Experiment 1 reveal an almost typical extinction pattern as well as the sensitivity of the EAMR to differential antecedent shock experience. The time rate of return to normal excretory cation levels differed for turtles given as little as 3 or 6 h of periodic antecedent shock.

The question of the discrepancy in stress-induced alkali metal excretion between reptile and rat, however, remains unresolved. The mere attribution of the discrepancy to phyletic difference is somewhat unsatisfactory in view of the evidence that the EAMR is present in man. It is possible, of course, that the conditioned emotional response—the procedure employed with rat—is not sufficiently stressful, or that the EAMR is itself insufficiently sensitive to either the qualitative or quantitative stress implicit in the CER experience. There is some evidence for the latter view in Paré's (1964) determinations that other physiological indices of CER-produced stress—those requiring sacrifice of the animal—were positive despite the failure to find increasing excretory cation concentrations in experimental Ss.

An even greater species or procedural difference appears to loom as a result of the data of Experiment 2. Turtles shocked in a random fashion over a period of 6 days showed rapid and clearly evident reduction of the EAMR. Ss administered the same amount of shock in a consistent daily pattern over the same period of time, however, revealed almost no extinction whatsoever of the EAMR during the subsequent nonshock days. The absence of any appreciable difference between levels of urinary Na^+ or K^+ between shock and nonshock for these Ss suggests that any extinction of the excretory cation response in nonrandomly shocked turtles would at the very least have been considerably prolonged. On none of the 10 subsequent nonshock days was there any overlap of mean urinary alkali metal levels for Ss systematically or randomly shocked during the preliminary 6-day treatment phase.

Although procedures radically differed, the current results are difficult to reconcile with the determination of chronic fear produced by unpredictable electric shock in rat (Seligman, 1968) or of the aversiveness of a series of shocks in humans being a function, in part, of uncertainty (Lovibond, 1968). Replication of the presently employed procedures with other species may help clarify the apparent discrepancies. For the present, however, it must be concluded that the stress indexed by extinction of the EAMR was considerably greater in a reptile previously shocked in a consistent rather than random manner.

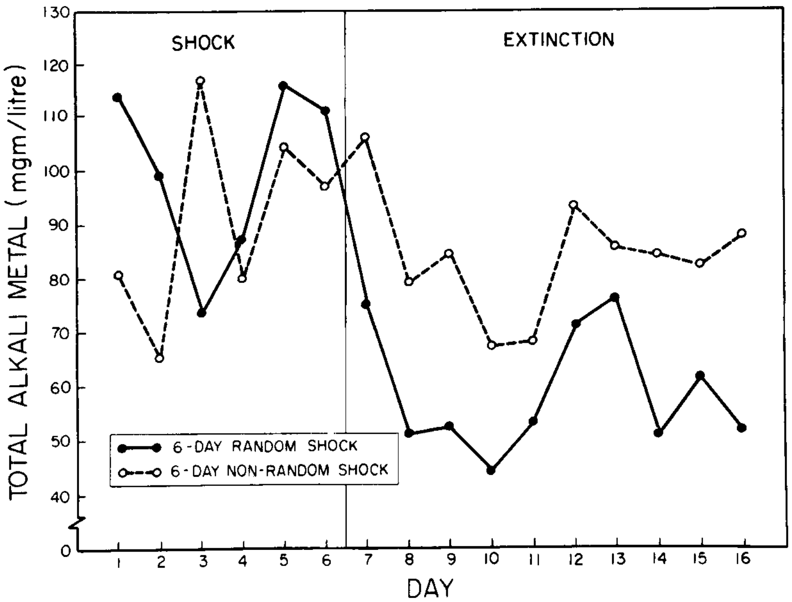

Fig. 2. Excretory alkali metal levels of Ss given a total of 48 h of periodic shock either systematically or randomly over a 6-day period and the subsequent urinary cation output for 10 subsequent nonshock days.

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NOTES

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2. Requests for reprints should be sent to Irwin M. Spigel, Department of Psychology, University of Toronto, Toronto 181, Ontario, Canada.

The effect of delay of punishment on choice behavior[1]

STEPHEN B. FLEISHMAN, Emory University, Atlanta, Ga. 30322, WILLIAM H. TEDFORD, JR., Southern Methodist University, Dallas, Tex. 75222, and CHARLES C. PERKINS, JR., Kansas State University, Manhattan, Kans. 66502

Twenty-four rats were trained to respond to two bars for reward delayed 30 sec. Shock was introduced contingent upon a response to the preferred bar during 120 choice trials during test. Group 1 received immediate shock, Group 2 received shock 24 sec after responding, and Group 3 received the 24-sec delayed shock, plus a signal between response and shock. Group 4 received Group 3 training plus 40 signal-shock pairings before testing. Response shift away from the preferred bar was significantly less in Group 2 than in the other groups, which were virtually indistinguishable.

The present experiment compared the effects of immediate punishment with three conditions of delayed punishment of a choice response. A choice response was selected for study because it is assumed that any general inhibition of activity from conditioned suppression, or a conditioned emotional response, affects pressing each of two bars equally. Thus, the only effect of the punishment on the choice measure is assumed to be weakening of the response tendency through a negative law of effect, the opposite of the reinforcing effect of a reward (Perkins, 1968).

Using a procedure which was modified only slightly for the present experiment, Tedford (1969) has shown that when there are mediating stimuli between the response and reward the specific (law of effect) suppressive effect of delayed punishment may be attenuated. Cairns (1969), using a similar procedure and a distinctive set of cues between the response and punishment, found that delay of punishment decreased the rate of change in choice but had no effect on percent choice at asymptote.

In the present experiment, one of the three delay groups had no external mediating stimuli between response and punishment. The other two delay groups had mediating stimuli between response and punishment. One of the latter two groups had received pretraining consisting of signal-shock presentations on which the warning signal was identical to the delay stimuli used subsequently.

METHOD

Data are reported from 24 naive Wistar rats between 130 and 300 g in weight. Half of the Ss in each group were males and half females. Six other Ss were discarded during the experiment because of E errors.

Standard Lehigh Valley apparatus employed included a two-retractable-bar test chamber (Model 1417F) with three jeweled lights over each bar, a Model 1531 constant-current shock source with scrambler, and a one-bar chamber. Other control and recording equipment was used as required by the fully automated procedure.

The Ss were reduced to 85% body weight and then shaped to barpress in the one-bar chamber. During this training and throughout the experiment, reward was .02 cc of 50% Borden's Eagle Brand sweetened condensed milk and 50% water, delivered by a dipper.

After the completion of preliminary training, all Ss were switched to the two-bar chamber and the delay of reward was gradually increased to 30 sec by a standard procedure. During this procedure, each block of three trials included one forced trial to each bar and one choice trial. At the end of this training, the sequence of events following a response to either bar was as follows: off-set of all lights, bars retract and dipper lowers into reservoir with audible click; 30 sec after the response, the houselight is turned on and the dipper starts to rise, taking 1.5 sec; 60 sec after the response, the bars are presented, the red jeweled lights above and to the right of each bar are turned on, and the houselight is turned off. Ss were then given a training day of 31 free-choice trials and were required to meet a criterion of completing the trials within 1 h with at least two responses to each bar. This criterion was usually met on the fifth day of training in the two-bar chamber.

Before the beginning of testing, Ss were assigned at random to one of four groups. The treatment for all Ss was the same on the 3 days of testing except for shock delay and associated cues. Shock was delivered following each response to the bar on which a majority of responses had been made on the criterion day. Shock was set for .3 mA and lasted .75 sec. On the first test day, each S received, in order, one forced trial to the preferred (shock) side, one forced trial to the nonpreferred side, and 40 free-choice trials. Forty choice trials were given on each of the following 2 days.

Group 1 received shock 1.5 sec after barpress, i.e., just as the bars had been completely withdrawn. For Group 2, shock was delayed by 24 sec. Group 3 had a 24-sec delay of shock and distinctive visual and auditory cues during the interval between barpress and shock. A pulsating

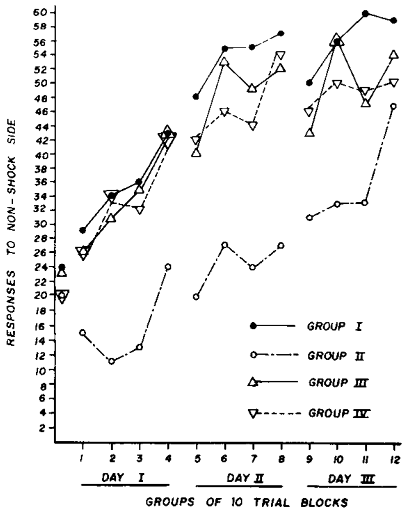

Fig. 1. Acquisition of preference to the originally nonpreferred bar for each group as a function of number of trials.