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Relapse of conditioned taste aversion in rats exposed to constant and graded extinction treatments



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ABSTRACT

In a generalized bait-shyness preparation of rats, the graded extinction procedure was not effective in preventing the relapse of conditioned aversion to a target taste. The present study is a replication of this finding in a conventional taste aversion preparation using a sodium chloride (NaCl) solution as the conditioned stimulus (CS) and an injection of poisonous lithium chloride (LiCl) as the unconditioned stimulus (US). After aversive conditioning of salty taste by a CS-US pairing, its extinction was administered by two different experimental procedures. The rats in the constant extinction group were repeatedly exposed to the target CS (NaCl solution). For the rats in the graded extinction group, the concentration of NaCl was gradually increased from a low level to the original one, and an additional interfering sweet taste was gradually faded out. The rats in the graded extinction group were equally or more prone to relapse than those in the constant extinction group. This result was observed by the second reacquisition treatment (Experiment 1), spontaneous recovery after a 30-day interval (Experiment 2), and renewal upon return to the acquisition context (Experiment 3). These results extended the generality of our previous finding, and its theoretical and clinical implications are discussed.

1. Introduction

One of the important findings of current conditioning research is the reappearance of conditioned behavior which is supposed to have been extinguished by post-conditioning exposure to the conditioned stimulus (CS) without the unconditioned stimulus (US). The reappearance occurs under several conditions, including the passage of time (spontaneous recovery), change of background contexts (response renewal), and reconditioning (reacquisition). These phenomena have been considered as animal models of symptom relapse after exposure therapies for maladaptive behavior (Bouton, 2000, 2002, 2004; Bouton & Nelson, 1998; Bouton & Swartzentruber, 1991; Bouton, Woods, Moody, Sunsay, & García-Gutiérrez, 2006; Laborda, McConnell, & Miller, 2011; Urcelay, 2012). Hence, any variables that might affect the occurrence and magnitude of these phenomena are worth exploring (e.g., Brooks & Bouton, 1993, 1994; Urcelay, Wheeler, & Miller, 2009).

In a recent article, we reported that a graded extinction procedure is equally or more susceptible to these response reappearance phenomena than the constant (*i.e.*, simple) extinction procedure. This finding was obtained in the generalized bait-shyness preparation with rats, wherein the avoidance of a salty taste was established by the voluntary ingestion of a salty and poisonous 0.15 M lithium chloride (LiCl) solution and then extinguished by the voluntary ingestion of a salty, but safe, sodium chloride (NaCl) solution (Nakajima, Onza, & Nakagawa, 2015). In the graded extinction procedure, a series of NaCl–sucrose compound solutions (*i.e.*, cocktails) were presented: the concentration of NaCl was gradually increased over days from a low level to 0.15 M and an additional

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Group	Acq	Extinction					Re	Test
Constant	★	★	★	★	★	★	★	★
Graded	★	☆	☆	☆	★	★	★	★

Fig. 1. The experimental design of the reacquisition experiment of Nakajima et al. (2015). Aversion to salty water was established by voluntary ingestion of 0.15 M LiCl solution (shown as a dark hexagram) on the acquisition day (Acq), and then it was extinguished over five days by the voluntary ingestion of 0.15 M NaCl solution (shown as a black pentagram) or gradually concentrated NaCl solutions (shown as light-to-dark pentagrams). After the reacquisition treatment (Re) with 0.15 M LiCl, conditioned aversion to salty water was assessed with 0.15 M NaCl. The initial adaptation training and days for recovery from illness were not shown for simplicity.

interfering sweet sucrose taste was gradually faded out. The rats in the constant extinction group, however, were allowed to drink 0.15 M NaCl solution throughout the extinction days. The critical assessment of conditioned salty aversion in these two groups was conducted with 0.15 M NaCl solution after a 23-day interval (spontaneous recovery), changing the background contexts (response renewal test), or reconditioning with 0.15 M LiCl intake (reacquisition). In all cases, the graded extinction rats were more prone to show aversion to the salty taste of NaCl solution than the constant extinction rats on the test day.

Nakajima et al. (2015) have discussed the vulnerability of the graded extinction procedure and its clinical implications; however, a full consideration of this procedure should be deferred until we have acquired more information about its generality. The present series of experiments thus extend the generality of the results of Nakajima et al. (2015) by employing the conventional conditioned taste aversion preparation, during which the poison was administered by an injection of LiCl. The target CS taste provided on the conditioning and test days is 0.15 M NaCl solution for all rats. The solutions employed in the extinction phase are the same as described by Nakajima et al.

Another reason to conduct the replication of Nakajima et al. (2015) with the conventional taste aversion preparation comes from our concern about the validity of the generalized bait-shyness preparation for assessment of relapse of conditioned aversion. Although the generalized bait-shyness preparation is ecologically more natural than the conventional taste aversion paradigm (Arriola, Alonso, Vázquez, & Rodríguez, 2015), it inevitably includes stimulus generalization between LiCl and NaCl solutions. Thus, one may ascribe any group difference observed in test performance to a difference in the extent of stimulus generalization rather than to the extent of relapse of aversion.

This point is especially important in the case of reacquisition, where the susceptibility to relapse is assessed by intake of 0.15 M NaCl solution after oral ingestion of a poisonous LiCl solution of 0.15 M (see Fig. 1). For the graded extinction rats, the 0.15 M NaCl and LiCl solutions are both presented once prior to reconditioning. For the constant extinction rats, on the other hand, the 0.15 M NaCl is repeatedly presented before the reconditioning. Accordingly, the two solutions are more *similar in familiarity* (i.e., number of presentations) for the graded extinction rats than for the constant extinction rats. Because stimulus generalization is greater when the stimuli are matched for familiarity (Honey, 1990; Robinson et al., 2017), generalized aversion from LiCl to NaCl should be greater in the graded extinction rats than in the constant extinction rats. The conventional taste aversion preparation, however, does not allow such reasoning, because stimulus generalization plays no role when the identical 0.15 M NaCl solution is employed in reconditioning and testing.

The experiments of the present study are reported in chronological order, starting with the assessment of reacquisition, followed by spontaneous recovery, and then renewal.

2. Experiment 1: reacquisition

Experiment 1 compares the ease of reacquisition after conducting the constant and graded extinction treatments in a conventional taste aversion preparation using an NaCl solution as the CS and an LiCl injection as the US.

2.1. Method

2.1.1. Subjects

A set of 16 experimentally naïve male albino rats (Slc:Wistar/ST) were purchased from the breeder (Japan SLC, Inc, Hamamatsu, Japan) at the age of 8 weeks and maintained on an *ad libitum* food schedule in individual hanging cages in a vivarium maintained at a temperature of 23 °C, a humidity of 55%, and a light/dark photoperiod of 16/8 h (the lights were switched on at 0800 h). The rats were tamed until they were 10 weeks old and their average body weights were 320.5 g (range: 290–340 g) on the first day of experimental treatment (Day 1).

2.1.2. Apparatus

The rats were carried in individual compartments of a cart between the vivarium and a quiet experimental room with 8 drinking cages placed on a table continuously lighted by the fluorescent lamps on the ceiling. These drinking cages were copies of their home cages, which were made of wire with two solid metal side walls (20 × 25 × 18.7 cm, w × l × h). Each rat consumed fluid from a glass bottle with a metal spout vertically inserted from the cage ceiling. The end of the spout was 14.5 cm above the cage floor.

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