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Short report

# Equating context conditioning in Pavlovian incubation of US signal value



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#### ABSTRACT

The present study replicated the finding by Goddard (2013), that when a single food pellet unconditioned stimulus (US) signaled the delivery of three additional pellets, responding after the single US was significantly greater on a delayed, compared to an early, test. Importantly, the present study equated context conditioning by delivering the identical number of food pellets in both conditioning and testing and showed that responding was significantly reduced in control subjects that had received unpaired single food–triple food presentations. Implications for theories of conditioning are considered.

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Typically, in Pavlovian conditioning, a neutral conditioned stimulus (CS), such as a tone, signals the delivery of a biologically potent unconditioned stimulus (US), such as shock. Given that certain conditions are fulfilled, organisms may learn this associative relation and display this learning with one or more conditioned responses (CRs).

In Pavlovian incubation, responding increases when Pavlovian conditioning is followed by a testing delay. For example, Pickens et al. (2009) found that following tone-shock pairings, conditioned fear in rats was significantly greater 31 and 61 days, compared to 2 or 15 days, following tone-shock training (see also McGaugh, 2003; Spear, 1978). A possible mechanism for Pavlovian incubation is that sleep consolidates mammalian memory, possibly through sleep-dependent transfer of recent memories from the hippocampus to neocortex (Marshall and Born, 2007; see also Both et al., 2004; Bunch, 1938; Bunch and Lang, 1939; Dijksterhuis and Nordgren, 2006; Dorfman et al., 1996; Erdelyi, 2010; Kamin, 1957; Yaniv and Meyer, 1987).

While Pavlovian incubation has been shown in fear conditioning, there is less evidence for Pavlovian incubation in appetitive conditioning (see Eysenck, 1979). However, a recent study by Goddard (2013) showed appetitive Pavlovian incubation in which

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a single food pellet signaled the delivery of three additional pellets. Previous studies from this lab have shown acquisition, extinction, spontaneous recovery, latent inhibition, blocking, renewal, and modulation, of US signal value, with important implications for conditioning theory (Goddard, 1996, 1997, 1999a,b, 2003; Goddard and Holland, 1996, 1997; Goddard and McDowell, 2001; Skinner et al., 1998).

Of most interest to the present study, however, was Experiment 1 in Goddard (2013). Using a within-subjects design, all rats first received four conditioning trials in which a single food pellet was followed, after 20s, by three additional pellets. Following conditioning, all rats then received testing in which a single food pellet was delivered and magazine entries were scored in the 20s interval after the pellet. All rats received two test sessions conducted one day after conditioning (Test 1), 14 days after conditioning (Test 2), 28 days after conditioning (Test 3), 42 days after conditioning (Test 4), and 56 days after conditioning (Test 5). Of most interest was that responding in subjects that had received four conditioning and four extinction test trials (Test 3) was significantly greater than the same subjects that had received only four conditioning trials (Test 1). This remarkable result provided the first demonstration of Pavlovian incubation in an appetitive conditioning preparation in which a single food pellet signaled the delivery of additional food.

However, note that in Goddard (2013, Experiment 1), subjects received four pellets in conditioning but only one pellet in testing. This may have reduced contextual associative strength in testing and there was some evidence of context extinction, as responding before the single food pellet showed modest declines in testing (see

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Goddard, 2013, Experiment 1, p. 106). Note that context extinction may have improved responding to the single food pellet, as suggested by Comparator Theory, because responding to a conditioning event may improve with a loss in the contextual comparator term (Miller and Matzel, 1988; see also Denniston et al., 2001). The present study addressed this problem by equating the number of pellets delivered in conditioning and testing. Specifically, a single food pellet was followed by three additional pellets after 20s (in conditioning) but not until after 2 min had elapsed (in testing). Thus, all subjects received four food pellets, in conditioning and testing, and the pellets were delivered in sessions of identical duration.

Note also that in Goddard (2013, Experiment 1), there was no control group that received unpaired single food–triple food presentations. This raises the possibility that increased responding in testing may have been an experimental artifact, possibly due to increased motivation in subjects experiencing more prolonged food deprivation. The present study addressed this problem by including controls that first received unpaired single food–triple food presentations. Response changes in testing could then be compared in subjects that had first received unpaired, as well as paired, single food–triple food presentations.

#### 1. Method

#### 1.1. Subjects

The subjects were 48 individually housed female Sprague-Dawley rats obtained from the Charles River Company. The subjects were about 130 days old at the beginning of the experiment and had not been involved in previous research. The animal holding room was maintained on a 14/10 h light/dark cycle with the lights on at 6:00 a.m. and off at 8:00 p.m. Subjects were maintained at 80% of their ad libitum weight, by measured feedings of Purina Rat Chow at the end of a session, and had continuous access to water in their home cages.

#### 1.2. Apparatus

The experimental chambers consisted of four identical boxes measuring  $28 \text{ cm} \times 21.5 \text{ cm} \times 21 \text{ cm}$ . The two side walls and top of each chamber were clear acrylic and the two end walls were aluminum. The floor consisted of 18 steel bars, each 0.5 cm in diameter, spaced approximately 1 cm apart. The front end wall contained a centrally located magazine, two lights, and a lever. The magazine was a dish-like structure, approximately 3.5 cm in diameter, that protruded from the wall at a height of about 2.5 cm from the floor. A single 45-mg dustless precision food pellet, manufactured by BIO-SERV (Holton Industries, Frenchtown, NJ), could be delivered into the magazine by a solenoid-operated feeder located behind the front end wall. The right and left lights were approximately 2 cm from the right and left edge of the front end wall, respectively, and 11.5 cm above the floor. Extending down from the center of the chamber was a 12 cm pole. Responses to the lever and pole were not recorded.

Each chamber was enclosed in a wooden shell containing an acrylic window permitting behavioral observation. A fan mounted inside each shell provided air circulation and a constant masking noise. Each shell also contained a houselight, mounted above the chamber, which consisted of six, 7.5-W, 120-V bulbs, passed through two layers of diffuse plastic to provide constant luminance.

Mounted approximately 20 cm outside each chamber was a video camera. In an adjacent room was a videocassette recorder, connected to each camera through a quad processor (National

Electronics, NLM Q4), and an IBM-compatible computer that controlled the four chambers.

#### 1.3. Procedure

Subjects first received one session of magazine training. Two food pellets were placed into the magazine (before subject placement) and subjects were left undisturbed for 10 min. After 10 min, two more pellets were delivered, followed by 10 additional pellets, separated by 1–2 min intervals in the remainder of the session. The houselights were then turned off to end the 30 min session. Following magazine training, subjects were randomly assigned to one of two groups with 24 subjects per group: Group Paired and Group Unpaired.

Conditioning followed magazine training. In Group Paired, subjects received a single food pellet delivered 10 min into each 14 min session. This single food pellet was then followed by three additional pellets 20s later. No other events were presented and the houselights were turned off to end the session. In Group Unpaired, subjects also received a single food pellet, delivered 10 min into each 14 min session, but this single food pellet was not followed by three additional pellets until after 2 min had elapsed. All subjects received 4 conditioning sessions (one session per day).

Following conditioning, all subjects then received testing in which a single food pellet was delivered 10 min into each 14 min session but was not followed by three additional pellets until after 2 min had elapsed. No other events were presented and the houselights were turned off to end the session. All subjects received two test sessions at regularly spaced intervals (one session per day) and responding was averaged over the two tests. These regularly spaced testing intervals included one day after conditioning (Test 1), 14 days after conditioning (Test 2), 28 days after conditioning (Test 3), 42 days after conditioning (Test 4), and 56 days after conditioning (Test 5).

#### 1.4. Data analysis

All observations were made from videotapes by a naïve observer blind to experimental conditions. Magazine entries were scored 20 s before, and 20 s after, the single food pellet delivered 10 min into each session. A magazine entry was defined as the placement of a rat's nose into the magazine (not including a placement to retrieve a food pellet). In addition, an inter-observer reliability check was conducted on test responding. While there was a correlation of .87 between the two observers, only the data from the naïve observer was used in all statistical calculations, although the data from both observers supported the same conclusions.

Throughout, data were evaluated by analysis of variance (ANOVA) with a rejection criterion of p < .05.

#### 2. Results

Before the single food pellet in conditioning, subjects in Group Paired and Unpaired responded at 1.9 (SEM=0.3) and 1.4 (SEM=0.2) magazine entries/min. This difference was not statistically significant (F(1, 46) = 2.1).

It was also of interest as to whether there was any evidence of temporal conditioning in Group Unpaired during conditioning; that is, could the subjects in Group Unpaired "anticipate" the arrival of the three food pellets that were delivered 2 min after the first food pellet? To check this possibility, magazine entries were scored in the 20s interval before the delivery of the three pellets. Subjects in Group Unpaired responded at 1.9 (SEM=0.5), 1.8 (SEM=0.6), 2.0 (SEM=0.5), and 1.4 (SEM=0.4) magazine entries/min during each of the four conditioning sessions, respectively. A One-Way Repeated Measures ANOVA showed no significant differences

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