



# Intertrial intervals and contextual conditioning in appetitive pavlovian learning: Effects over the ABA renewal paradigm



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## ABSTRACT

Three experiments using rats in an appetitive conditioning procedure analyzed the effect of short and long (50 s vs. 1440 s) intertrial intervals (ITI) over the acquisition of conditioned stimulus (CS), context (Ctxt), and unconditioned stimulus (US) associations, as well as the effect on the extinction and renewal of the conditioned response to the CS. Experiment 1 revealed more contextual conditioned responses in groups trained with the short ITIs, however the renewal effect was not observed during test phase with either ITI condition. When subjects were pre-exposed to the contexts before the acquisition phase (Experiment 2) renewal of the conditioned response (CR) was only observed in long ITI group. However, when the acquisition context was extinguished (Experiment 3) the renewal effect observed in the Experiment 2 was weakened. In all three experiments subjects showed a similar number of responses to the tone predicting food, however they showed a clear contextual conditioning effect only for the groups trained with short ITIs. It is noteworthy that the acquisition context showed high levels of the CR in the renewal test only for groups trained with short ITIs (Experiment 2) but these responses were absent if additional contextual extinction was imposed before such test (Experiment 3). In general, all groups showed similar acquisition curves for the CS but only Short groups had an increase in the CR during the pre-CS. Also, context conditioning does not interfere with the conditioning of the CS and context pre-exposure prior to acquisition is essential in order to observe the renewal effect when long ITIs are used.

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## 1. Introduction

Nowadays there is a growing interest in understanding how contextual stimuli influence behavior (e.g. Urcelay and Miller, 2014; Holland and Bouton, 1999; Maren et al., 2013), since it has been observed that these stimuli plays a key role in associative learning. The intertrial interval (hereafter, ITI) has been one of the most studied variables when contextual learning is analyzed.

The importance of the ITI length over the development of the pavlovian conditioning has been pointed out by several researchers (Barela, 1999; Gibbon and Balsam, 1981; Miller and Matzel, 1988; Rescorla and Wagner, 1972; Wagner, 1981). For example, Rescorla and Wagner (1972) suggested that the ITI length plays an important

role in the strength of the conditioned stimulus (CS) and the unconditioned stimulus (US) associations, as well as the Context–US associations. These authors propose that during the conditioning trials, two associations are strengthened: one between the CS and the US, and other between the Context and the US, because both stimulus (i.e. Context and CS) are present when the US is delivered at the end of the trial. However, during the ITI, Context–US associations are weakened (i.e. extinction) because the context is the only stimulus present during these intervals, and no reinforcers are presented during the ITI. Accordingly, short ITI lengths result in more contextual conditioning, while long ITI lengths result in less contextual conditioning (e.g. Terrace et al., 1975). Additionally, it is expected that the associative strength of the CS be affected by this Context–US association, thus with short ITIs there is more competition between the CS and the Context for associative strength, and with long ITIs the CS acquire the associative strength easily because context's associative strength is extinguished during these intervals. For example, Rescorla and Durlach (1987) showed in a within-subjects autoshaping experiment with pigeons as subjects, that ITIs produced greater conditioning to a CS trained with long

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ITI, as well as, a context having short ITIs showed greater ability to promote responses to stimuli trained in any other context.

Sunsay and Bouton (2008) conducted a similar experiment with rats trained in an appetitive conditioning procedure. In their Experiment 1, they exposed eight groups of rats during 16 acquisition sessions to four daily trials where a 30 s tone was paired with food pellets; each group was conditioned using a different ITI length (i.e. 60, 120, 180, 240, 480, 960, 1440 and 1920 s). The food-cup entries during the CS and during the 30 s before the CS (i.e. pre-CS or pre-period) were registered as CRs to the CS and to the context, respectively. Acquisition showed a decreasing tendency in the amount of food-cup entries during the pre-CS as the ITI length increased, but the CR to the tone was relatively constant across all the different ITI lengths. The authors concluded that the decrease in the response level in the pre-CS period reflects the decrease of the Context–US associative strength. These results were consistent with the Rescorla and Wagner (1972) model's prediction regarding the extinction of the Context–US association during the ITI, thus as the ITI length increases, more contextual extinction should be observed. In conclusion, the length of the intertrial interval affects the strength of the CS and context conditioning. Most findings suggest that long intertrial intervals results in high conditioning to the CS, and short intertrial intervals produce high contextual conditioning. This evidence comes to relevance in several contextual phenomena, particularly in the ABA context renewal paradigm.

The context renewal effect is usually observed in procedures arranged in three phases: Acquisition, Extinction and Test. Particularly, in the ABA renewal design the acquisition phase is conducted in Context A, the extinction phase occurs in Context B and the test phase keeps the same conditions of extinction but the subjects return to the acquisition context (Bouton and Peck, 1989; Bouton and Ricker, 1994; Brooks and Bouton, 1994). Additionally, contextual renewal has been observed when the acquisition and extinction contexts are the same, while test context is different (i.e. AAB renewal), as well as when the contexts used in each phase are different (i.e. ABC renewal). To account for these observations, Bouton (1993) suggested that the context renewal effect is observed when extinction treatment is given in a different context from that used during the extinction phase. Thus, context renewal is expected when extinction and test contexts differed. Also, it is assumed from Bouton's explanation of the renewal effect that the level of response recovery should be similar in all designs, however findings using very different preparations (e.g. instrumental conditioning, fear conditioning) have failed to support this prediction (e.g. Nakajima et al., 2000; Thomas et al., 2003), in fact it has been observed higher levels of context renewal when the ABA renewal design was used. In agreement some authors have suggested that some contextual conditioning could be observed during the acquisition (Lovibond et al., 1984; c.f. Bouton and King, 1983). For example, some experiments have showed a reduction in the level of response when acquisition context is changed after the conditioned response has been acquired. Additionally it has been observed that the context and the conditioned stimulus compete in order to acquire associative strength and this could be affected, among several factors, by the length of the intertrial interval.

Since the ABA design is the only renewal design that uses the acquisition context in the test phase, it is reasonable to assess whether a higher response recovery in this design could be produced by an interaction between the mechanism proposed by Bouton (i.e. context change between extinction and test) and the associative strength of the acquisition context. Bouton and King (1983) tested this possibility in four conditioned suppression experiments. In these experiments, they found that the ABA renewal effect does not depend on contextual conditioning of the test context but still they were able to find contextual conditioning of the acquisition context and they were not able to determine

if such contextual conditioning played a role in the renewal effect. In order to test these assumptions in a pavlovian appetitive procedure, we designed three experiments to assess if the training ITI length may affect contextual conditioning, and if this contextual conditioning may increase ABA renewal. Particularly, we assessed if subjects trained with short ITI lengths show a higher renewal effect than those trained with long ITIs.

## 2. Experiment 1

In the first experiment, four groups of rats were trained in an appetitive conditioning procedure where a 30 s tone was paired with food. All groups were trained during six sessions in Context A, in two groups (i.e. ABA Short, AAA Short), trials were spaced by short ITIs (i.e. 50 s), whereas in the other two groups (i.e. ABA Long, AAA Long) long ITIs (i.e. 1440 s) were used. Then all groups underwent extinction. AAA Short and AAA Long groups received extinction trials in Context A, while ABA Short and ABA Long received extinction in Context B. Finally, all subjects were tested in the context where training occurred. During the last two phases of the experiment, all groups were extinguished and tested using an intermediate ITI length (i.e. 275 s). This design allowed us to assess the effect of contextual conditioning in acquisition, extinction and contextual renewal. Since AAA Short and ABA Short groups were trained with short ITI lengths we expected to observe high levels of contextual conditioning, while in AAA Long and ABA Long low levels of contextual conditioning were anticipated (e.g. Denniston et al., 2003; Mustaca et al., 1991; Rescorla and Durlach, 1987; Sunsay and Bouton, 2008; Urcelay and Miller, 2010). Additionally, in this experiment we omitted the pre-exposure phase described in most appetitive classical conditioning renewal experiments (Bouton et al., 1993; Bouton and Peck, 1989; Brooks and Bouton, 1994; Ricker and Bouton, 1996), in order to avoid a possible interaction between latent inhibition of the context and ITI length, as it has been observed (at least in aversive conditioning procedures) that context pre-exposure results in latent inhibition to the context (Cole et al., 1996; Urcelay and Miller, 2010).

### 2.1. Method

#### 2.1.1. Subjects

The subjects were 48 experimental naïve Wistar rats (6 male and 6 female per group) bred at the Psychology school vivarium (Universidad Nacional Autónoma de México). They were approximately 90 days old and weighing 200–350 g at the beginning of the experiment and were housed individually in standard cages, in a colony room. The experiment was conducted on consecutive days during the light portion of the day. The rats were food deprived and maintained at 83% of their free-feeding weights throughout the experiment, they also had free access to water on their individual Plexiglas home cages and at the end of every session they received complementary food to keep them on their deprivation goal weight.

#### 2.1.2. Apparatus

Experimental sessions were conducted in six MED Associates® conditioning chambers measuring 20.8 × 21 × 28.2 cm (*h × l × w*). Each chamber consisted of a front and a rear stainless steel panel, while the superior panel and sidewalls were made of clear acrylic sheets. The floor of all chambers consisted on 16 tubular stainless steel bars, of 0.5 cm diameter, separated by a 1.5 cm center-to-center gap and which were located parallel to the front panel. In the center of the front panel and 1 cm above the floor, there was located a 5 cm × 5 cm quadrangular opening and inside of it a food-cup was located. Behind the front panel was a pellet dispenser, which delivered 45 mg Bio-Serv Dustless Precision Pellets®, rodent grain based diet (Bio-Serv, Frenchtown, NJ). The US was the presentation of two 45 mg Bio-Serv precision pellets

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