

ABA, AAB and ABC renewal in taste aversion learning

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Context renewal is identified when the conditioned response (CR) elicited by an extinguished conditioned stimulus (CS) reappears as a result of changing the contextual cues during the test. Two experiments were designed for testing contextual renewal in a conditioned taste aversion preparation. Experiment 1 assessed ABA and AAB context renewal, whereas Experiment 2 assessed ABA, AAB and ABC context renewal. The procedure of both experiments included three successive phases: acquisition, extinction and testing. The acquisition phase involved one pairing of sucrose flavor with a lithium chloride (LiCl) injection in Context A for all groups of rats. The groups were exposed to the sucrose flavor alone during the extinction phase, for the groups AAB and AAA extinction was carried out in Context A, whereas groups ABA and ABC were extinguished in Context B. Testing was done in context A for the groups ABA and AAA, whereas group AAB was tested in context B; lastly, the ABC group was tested in Context C. Results showed ABA, AAB and ABC renewal of conditioned taste aversion. The results are interpreted and discussed in relation to Bouton's (1994) retrieval of information model.

In an extinction procedure conditioned responding (CR) diminishes when a conditioned stimulus (CS) is presented in absence of the unconditioned stimulus (US); however, if the CS is presented in a different array of contextual cues, such as a different chamber, the CR reappears, this effect is known as context renewal. In a context renewal procedure the CS-US association is established in one context (i.e. Context A), extinguished in a different one (i.e. Context B), and finally the test session is conducted in the acquisition context (ABA renewal, e.g. Rosas & Bouton, 1997).

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Renewal has also been demonstrated when acquisition, extinction and testing are conducted in three different contexts (ABC renewal, e.g. Bouton & Bolles, 1979) or when acquisition and extinction take place in the same context but testing occurs in a different one (AAB renewal, e.g. Bouton & Ricker, 1994).

Bouton (1993, 1994, 1997) proposed the retrieval of information model to account for renewal and other retrieval-related phenomena such as spontaneous recovery, reinstatement, etc. According to this model, conditioning generates memories of the target events that become associated to each other. During acquisition, associations between the memories or nodes of the CS and US are established, thus when the CS is presented it activates its representing node as well as the US node to which it is associated. It is assumed that non-reinforced presentations of the CS create a new inhibitory association between the CS and US nodes during extinction. Thus, after extinction, the CS node has two associations with the same US node, one excitatory and other inhibitory. The model proposes that activation of the inhibitory association is modulated by contextual extinction cues. That is, the contextual cues presented during extinction activate an intermediate node that is assumed to possess AND gate properties (i.e. as an occasion setter); that is, when the CS and context nodes are simultaneously active the inhibitory association prevents the full activation of the US node and thus a performance similar to extinction is observed (Bouton, 1993; Bouton & Ricker, 1994). On the other hand, if the context node is not active at the same time as the CS node, the excitatory association will cause the US node to be active producing renewed responding. In summary, the retrieval of information model predicts renewal when test session is conducted in a different context from the extinction context; therefore the three renewal designs would be nominally identical.

However, findings reported in the literature do not support this prediction of the model. For example, Thomas, Larsen and Ayres (2003) conducted a direct comparison between AAB, ABA and ABC renewal with a fear conditioning procedure and found that the size of AAB renewal was smaller than the size of the other two (cf. Rescorla, 2008). Sánchez-Carrasco, Bernal-Gamboa and Nieto (in preparation) found a similar result with an appetitive instrumental conditioning procedure. Moreover, Harris, Jones, Bailey and Westbrook (2000) demonstrated larger ABA than ABC renewal after extinction in an aversive conditioning procedure. Furthermore, Üngör and Lachnit (2008) conducted an experiment with a predictive learning task in humans and found ABA and ABC renewal, but no AAB renewal (see also Goddard, 1999; Nakajima, Tanaka, Urushihara & Imada,

2000). There is also evidence that AAB renewal is harder to find than ABC or ABA renewal (Rosas, Callejas-Aguilera, Ramos-Alvarez & Abad, 2006; Tamai & Nakajima, 2000). In conclusion, a significant difference has been observed between the size of AAB, ABA and ABC renewal, which seems to depend of the learning procedure used. To account for these differences Rosas et al. (2006) have suggested that ABA and ABC renewal involve additional mechanisms to the context change between extinction and testing. Therefore, the analysis of the factors that produce the differences observed between the size of AAB, ABA and ABC renewal could provide some information about these mechanisms. Particularly, Rosas and Bouton (1997) found ABA renewal with a conditioned taste aversion (CTA) procedure, whereas Rosas, García-Gutiérrez and Callejas-Aguilera (2007) reported AAB and ABA renewal. Since to our knowledge, ABC renewal has not been reported in CTA the present experiments sought to provide such evidence. In addition, they had two goals: First, they are a systematic replication of the ABA and AAB renewal effect reported by Rosas et al. (2007); and secondly, they compared directly ABA, ABC and AAB renewal as a way of testing Bouton's (1993) predictions of renewal.

EXPERIMENT 1

The goal of Experiment 1 was to observe the ABA and AAB renewal effect reported by Rosas and Bouton (1997), and Rosas et al. (2007). This was relevant because experiments conducted in our laboratory failed to produce taste aversion learning using the 0.15M dose, at 2% body-weight, of LiCl as the US used by Rosas et al. (2007); other researchers have also encountered difficulties in reliably producing the renewal effect (Gallo, personal communication). In the present experiments the LiCl dose was increased to .3M, at 0.5% of body-weight (Holland, 1981). An additional difference was that, we used sucrose instead of saccharine flavor as the CS, and in order to increase the differences between contextual cues (v. Thomas et al., 2003) we added an odor cue to the contexts described by Rosas et al. (2007).

Three groups of rats (AAA, ABA and AAB) received a single conditioning trial where sucrose consumption was paired with an injection of LiCl in Context A. Groups AAA and AAB received three extinction trials in Context A, while group ABA received the extinction trials in Context B. Groups AAA and ABA were then tested in Context A, while group AAB was tested in Context B. In order to equate the subject's familiarity with both contexts, rats received two daily sessions throughout the experiment, one in Context A, and the other in Context B (see Rosas &

Bouton, 1997; Rosas, et al., 2007). In one of these sessions, rats were given plain distilled water and liquid sucrose in the other.

METHOD

Subjects. Twenty-four female Wistar rats weighing in average 269 g were used. They had been previously trained to lever press to obtain food for 30 days. At the end of that training, all subjects were maintained with ad libitum access to food and water in their home cages for two weeks and their weights were re-determined. Twenty-four hours before the beginning of the experiment, subjects were water deprived with free access to food in their home cages. During the experiment subjects had access to one of the fluids for 15 min every 12 h, the first session occurred at 7:00 h and the second at 19:00 h.

Apparatus. Eight Plexiglas cages measuring 22 x 20 x 43 cm were set up to provide two different arrays of contextual cues. The walls of four cages were covered with dark green sheets of paper and the floor was covered by recycled fiber paper egg trays adapted to the floor of each cage; a cotton wool scented with 10 ml of white vinegar (Clemente Jacques, Sabormex S.A. de C. V., México) was placed under the paper egg tray of each cage. The walls of the four remaining cages were covered with white paper with red squares of 7 mm in width, the floor of each cage was covered by perforated chipboard adapted to it, a cotton wool scented with 10 ml of anise (McCormick & Company Inc., Maryland) was placed under the perforated chipboard. These arrays of contextual cues were counterbalanced across groups as Context A or B.

The CS was liquid sucrose at 15% (w/v) and was provided to the rats in a 50 ml graduated cylinder with an adapted sipper tube. The US was a single i.p. injection of LiCl, at .3M, in a dose of 0.5% of bodyweight.

Procedure. The design used in this experiment is schematically shown in Table 1. During the first three days subjects received two 15 min sessions of free access to tap water in their home cages and were assigned to groups ABA, AAB and AAA by matching their water consumption in these sessions. The first drinking session started at 07:00 h and the second one occurred at 19:00 h (“morning” and “evening” drinks, respectively).

In the next two days, subjects were exposed to the contexts in a fully counterbalanced manner in two daily sessions. They were divided into two groups matched on morning and evening consumption. One of groups was

assigned to one array of boxes in the morning and the other in the evening; while, the other group received reverse assignment. This procedure was maintained throughout the experiment, and made it possible to consider time of day of deprivation and boxes as part of the context. Rats were assigned to one box/time as Context A and to the other as Context B. On the first session they were exposed to one context and to the alternative context on the second session. On each of these sessions they received 15 min of free access to tap water.

On the sixth day subjects received the CTA acquisition trial. On this trial, all groups had 15 min access to sucrose in Context A, subsequently the drinking tube was removed and 10 min were allowed to elapse. Then, each subject was given an i.p. injection of LiCl and was returned to Context A for an additional 15 min period. At the end of this session subjects were returned to their home cages. In the alternative daily sessions rats were simply given access to tap water for 15 min in Context B. The following day, subjects were given the two sessions of free access to water (15 min each) in their home cages. On days eight to ten, extinction trials were conducted. All groups received access to sucrose in one session and water in the other session, group ABA was extinguished in Context B, whereas groups AAB and AAA were extinguished in Context A.

Renewal test was conducted on the eleventh day. As in the previous phases, all subjects had access to sucrose in one session and water in the other session. Groups ABA and AAA received sucrose in context A, while group AAB was tested in context B.

Statistical analyses. Liquid consumption was recorded to the nearest milliliter throughout the experiment. Consumption was compared using analyses of variance (ANOVA), and planned comparisons were made using the methods proposed by Maxwell and Delaney (2003).

RESULTS AND DISCUSSION

Acquisition. The left-hand panel of Figure 1 shows sucrose consumption during the acquisition trial. Consumption was compared using a one-way ANOVA, which showed that groups did not differ, $F(2, 21) = 1.68$, $MSE = 10.30$, $p > 0.05$.

Extinction. The center panel of Figure 1 shows the mean sucrose consumption for each group during extinction trials. Sucrose consumption on the first extinction trial was reduced for all groups revealing a strong

aversion for sucrose, and as extinction continued sucrose consumption increased. A two-factor ANOVA including Group (3) and Trial (3), with Trial as repeated measures, confirmed a significant effect of Trial, $F(2, 42)=83.22$, $MSE = 2.26$, $p < 0.05$, but the main effect of Group, $F(2, 21)=0.26$, $MSE=6.09$, $p > 0.05$ and the interaction Group x Trial were non significant, $F(4, 42)=0.47$, $MSE=5.99$, $p > 0.05$. Thus, extinction proceeded as expectedly, with the groups showing a similar and decreasing level of taste aversion, at the beginning and the end of extinction.

Test. The right-hand panel of Figure 1 shows the mean sucrose consumption per group during the test trial. It can be noticed that groups ABA and AAB drank less than the control group AAA, these differences were significant, $F(2, 21) = 6.55$, $MSE= 12.00$, $p < .05$. Planned comparisons of the groups showed that the magnitude of renewal was similar for groups ABA and AAB since they did not differ, $F(1,21) = 0.62$, $MSE= 7.5$, $p > .05$. On the other hand, groups ABA and AAB both differed significantly from group AAA, $F(1, 21) = 12.48$, $MSE= 12.05$, $p < 0.05$, respectively.

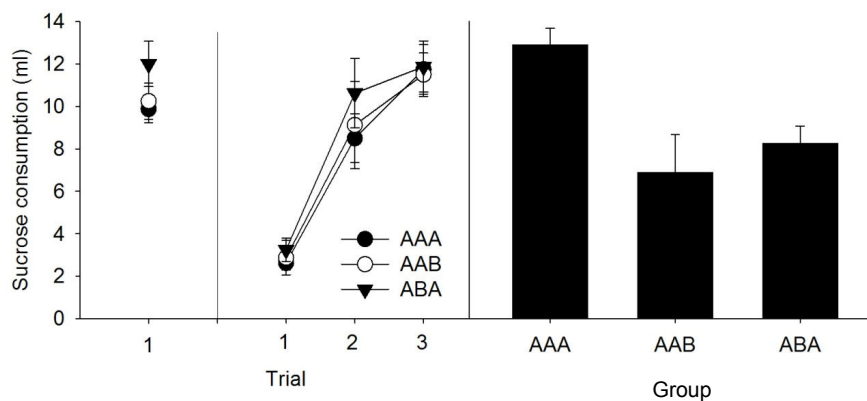


Figure 1. Mean sucrose consumption in ml in Experiment 1, during the single acquisition trial (left panel), on the three extinction trials (center panel), and during the test trial (right panel) for the ABA, AAB and AAA groups.

In summary, the present results replicate the ABA renewal effect using a CTA procedure reported by Rosas and Bouton (1997) and the AAB renewal effect reported by Rosas, et al. (2007) with a modified procedure. Similarly, these results showed that the size of ABA and AAB renewal is similar when three extinction trials are used.

EXPERIMENT 2

The results of Experiment 1 are consistent with previous demonstrations of ABA (Rosas & Bouton, 1997) and AAB renewal (Rosas, et al., 2007) with a CTA procedure. Furthermore, they may be interpreted as support to Bouton's retrieval information model (1993, 1994 & 1997). According to this model, renewal should be observed when the meaning of the CS changes while extinction take place, so that extinction becomes context specific. Thus, the size of the AAB, ABA and ABC renewal should be identical, since in the three procedures there is a context change after extinction.

The second goal of these experiments was to conduct a direct comparison of ABA, ABC and AAB renewal. As we mentioned earlier Bouton's retrieval of information model (1993, 1994 & 1997) propose that the three types of renewal design should produce the same magnitude of renewal. However, there are some findings in the literature that do not support this prediction (Thomas, et al., 2003; Üngör & Lachnit, 2008). Particularly, some studies report larger ABA than ABC renewal in rats (Harris, et al., 2000; Bouton & Brooks, 1993; Rescorla, 2008) and humans (Üngör & Lachint, 2006). Thus, in Experiment 2 we directly compared the magnitude of AAB, ABC and ABA renewal.

In this experiment four groups of rats (AAA, ABA, ABC and AAB) were exposed to three daily sessions one for each context (A, B and C), in order to match the contexts familiarity or even their associative strengths. In two of these consecutive sessions rats were given plain distilled water and liquid sucrose in the other. The four groups of rats received a single conditioning trial where sucrose consumption was paired with an injection of LiCl in Context A. Groups AAA and AAB received three extinction trials in Context A, while groups ABA and ABC received the extinction trials in Context B. Groups AAA and ABA were then tested in Context A, group AAB was tested in Context B and group ABC was tested in Context C.

METHOD

Subjects and Apparatus. Forty-eight naive females Wistar rats weighing 293 g in average were used, but one was excluded from the experiment due to sickness. The rats were assigned to one of the four groups (N=12 rats in each group, except for the AAA group, N=11) according to their mean water consumption in the three days previous to acquisition. They were maintained in the same conditions described for Experiment 1, except that rats had access to water three times a day (0900, 1400 and 1900h, “morning”, “afternoon” and “evening” drinks, respectively), for 10 min.

The two different sets of Plexiglas described in Experiment 1 were used in this experiment, and a new context was included, being the three different contexts counterbalanced among subjects. The four additional Plexiglas cages (18 x 15.5 x 28 cm) had their walls covered with a diagonal pattern paper (black bars 0.6 cm wide and white bars of 0.8 cm wide). The floor was covered with foam sheet adapted to each cage and under it a piece of cotton scented with 10 ml of Windex (S.C. Johnson and Son, S.A. de C.V. México) was placed.

Procedure. The procedure was identical to that described in Experiment 1, except that in this experiment there were three 10 min daily sessions (0900, 1400 and 1900h), one for each available context. The acquisition trial of CTA for all groups was conducted in Context A; this consisted of a single pairing of sucrose consumption with an i.p. injection of LiCl as described earlier; extinction of taste aversion was conducted in Context B for groups ABA and ABC, and in Context A for groups AAA and AAB, all groups were given three sessions in which drinking sucrose was not paired with LiCl; finally, the group ABC was tested in extinction in Context C, group AAB was tested in Context B and groups AAA and ABA were tested in Context A (see Table 1).

RESULTS

Acquisition. The left-hand panel of Figure 2 shows the mean sucrose consumption in ml during the acquisition trial for each group. A one-way ANOVA test showed that the sucrose consumption did not differ among groups, $F(3,43) = 0.66$, $MSE = 5.83$, $p > 0.05$.

Extinction. The center panel of Figure 2 shows sucrose mean consumption for each group during the three extinction trials. Sucrose

consumption on the first extinction trial was reduced for all groups revealing a strong aversion for sucrose and as extinction continued sucrose consumption increased. A two-factor ANOVA with repeated measures in one factor (three trials) showed that only main effect Trial was significant, $F(2, 86) = 98.43$, $MSE = 407.2$, $p < 0.05$, but neither Group $F(3, 43) = 0.26$, $MSE = 6.1$, $p > .05$, nor the interaction Group \times Trial were significant, $F(6, 86) = 0.18$, $MSE = 0.80$, $p > .05$.

Table 1. Design of Experiments 1 and 2, and groups used in each experiment. Characters before colon show the context in which that phase of the experiment was conducted. Numbers correspond to the number of trials conducted on each phase. S stands for sucrose, + for LiCl injection, and – for the absence of injection.

Experiment	Group	Acquisition	Extinction	Test
1	AAA	A: 1S+	A: 3S-	A: 1S-
	AAB	A: 1S+	A: 3S-	B: 1S-
	ABA	A: 1S+	B: 3S-	A: 1S-
2	AAA	A: 1S+	A: 3S-	A: 1S-
	AAB	A: 1S+	A: 3S-	B: 1S-
	ABA	A: 1S+	B: 3S-	A: 1S-
	ABC	A: 1S+	B: 3S-	C: 1S-

Test. The right-hand panel of Figure 2 shows the mean consumption for each group during the test. It can be seen that groups ABA, AAB and ABC drank less sucrose ($M = 5.91$ ml [$SE = 0.95$], $M = 5.50$ ml [$SE = 0.95$], $M = 6.75$ ml [$SE = 0.95$]) than group AAA (10.36 ml [$SE = 0.99$]) in the test trial; these differences were significant, $F(3, 43) = 5.06$, $MSE = 55.37$, $p < .05$. Planned comparisons of the groups showed that the magnitude of renewal was similar for groups ABC against ABA and AAB, $t(43) = 0.89$, $SE = 3.41$, $p > 0.05$. On the other hand, groups ABA, ABC and AAB differed significantly from group AAA, $t(43) = 3.80$, $MSE = 3.41$, $p < 0.05$.

In summary, the present results showed that the size of ABA, AAB and ABC renewal did not differ when three extinction trials are used. They also contradict previous reports (Thomas, et al., 2003; Üngör & Lachnit, 2008) and support the prediction of Bouton's retrieval of information model.

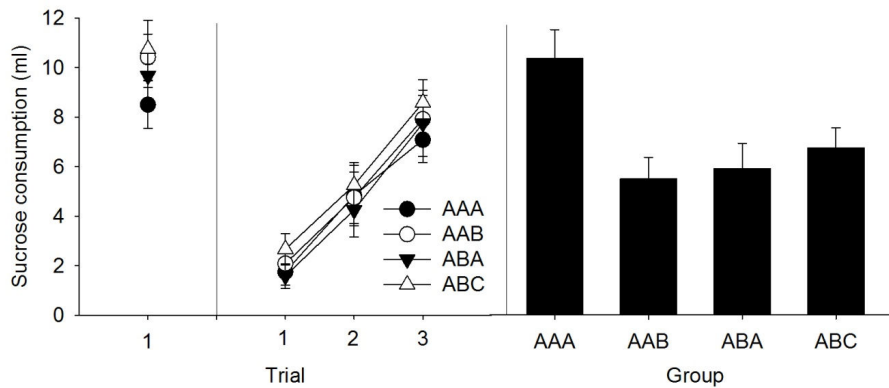


Figure 2. Mean sucrose consumption in ml in Experiment 2, during the single acquisition trial (left panel), on the three extinction trials (center panel), and during the test trial (right panel) for the AAA, AAB, ABA and ABC groups.

GENERAL DISCUSSION

The experiments reported here explored ABA, AAB and ABC renewal in a CTA procedure. Results of Experiment 1 showed ABA and AAB renewal, a result that is consistent with earlier published reports (e.g. Rosas & Bouton, 1997; Rosas, et al., 2007). Experiment 2 was able to demonstrate ABC renewal, a result that extends the range of phenomena observed with this procedure. Furthermore, this experiment showed that ABA, AAB and ABC renewal designs produce similar magnitudes of renewal. Consequently, the present results support a central claim of Bouton's (1994) retrieval of information model, according to which similar magnitudes of renewal should be observed when testing is conducted in a different context from the one used in extinction. However, there are some published findings that are inconsistent with this prediction (Bouton & King, 1983; Harris, et al., 2000; Rosas, et al., 2007). For instance, Thomas, et al. (2003) using a fear conditioning procedure reported that AAB renewal was smaller than ABA and ABC renewal, which did not differ between them. A similar result

was reported by Üngör and Lachnit (2008) with a reversal learning procedure with humans. Conversely, Rosas, et al. (Experiment 1, 2007) showed that ABA and AAB renewal were similar in a CTA procedure; while Harris, et al. (2000) reported that ABA renewal was larger than ABC renewal. In this regard, the similarity in magnitudes of renewal found in the present experiments may reflect a measure of renewal on a changing continuum of extinction trials, since there is evidence showing that AAB and ABA renewal are differentially affected by increasing the number of extinction trials. For example, Rosas, et al. (2007) have shown in a CTA procedure that AAB renewal disappears as the number of extinction trials increase (from 3 to 5 trials), a similar result was reported by Tamai and Nakajima (2000) using a fear conditioning procedure (72 trials to 112 trials). Similarly, Denniston, Chang and Miller (2003) found attenuated ABA and ABC renewal after 800 extinction trials but not after 160 trials. Clearly, these findings do not support Bouton's model prediction (1993) that increasing the number of extinction trials should also increase the value of context as an occasion setter; that is, extinction should become increasingly more context specific as the number of extinction trials increase. It is necessary to further analyze the effects of the length of extinction (i.e. number of trials) on the magnitude of renewal, as well as other explanations. For instance, Denniston, et al. (2003) have suggested that increasing extinction training increase the generalization gradient of the CS - No US association to contexts different from the extinction context.

In conclusion, although our findings support the retrieval of information model (Bouton, 1993, 1994 & 1997) in that the magnitude of renewal should be similar, there is a growing set of data suggesting that the mechanisms underlying the three renewal designs may be different to those proposed by Bouton (1994), and could include the length of extinction which may influence the different magnitudes of the renewal effect.

RESUMEN

Renovación ABA, AAB y ABC en condicionamiento de aversión a sabores. Existe evidencia de que los cambios en los estímulos ambientales afectan activamente la ocurrencia de la respuesta condicionada (RC). La renovación contextual o la reaparición de una RC extinguida se observa cuando se continúa la extinción de dicha respuesta en un contexto diferente. Se diseñaron dos experimentos con el propósito de evaluar la renovación contextual en un procedimiento de condicionamiento de aversión al sabor. En el Experimento 1, se analizó la renovación contextual ABA y AAB, mientras que el Experimento 2 se evaluó la renovación ABA, AAB y ABC. Ambos experimentos constaron de tres fases: adquisición, extinción y

prueba. En ambos experimentos, la fase de adquisición se condujo en el Contexto A para todos los grupos y en ella, se inyectó i.p. Cloruro de Litio (LiCl) a los sujetos inmediatamente después de que ingirieron una solución de sacarosa. En la fase de extinción, se expuso a los sujetos únicamente a la solución de sacarosa, sin la inyección. Los grupos AAB y AAA recibieron esta fase en el Contexto A, mientras que los sujetos de los grupos ABA y ABC la recibieron en el Contexto B. En la fase de prueba, se continuó la extinción. Los grupos ABA y AAA recibieron esta fase en el Contexto A, el grupo AAB en el contexto B, mientras el grupo ABC la recibió en un tercer contexto. Los resultados mostraron renovación de la aversión condicionada al sabor en los grupos ABA, AAB y ABC. Se analiza la relación de estos hallazgos con el modelo de recuperación de información de Bouton (1994).

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