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Illness-Induced Context Aversion Learning in Rats With Lesions of the Dorsal Hippocampus

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In 2 experiments, rats with electrolytic lesions of the dorsal hippocampus and sham-operated control subjects were given injections of lithium chloride after exposure to a distinctive context. This procedure establishes a context–illness association in intact subjects. In Experiment 1, the strength of the context aversion was assessed by measuring the subjects' willingness to consume a novel flavor in the context. It was found that lesioned subjects showed less suppression of consumption than controls. Experiment 2 tested the ability of the context to block subsequent flavor-aversion learning and revealed less effective blocking in lesioned rats. These results are consistent with the view that hippocampal lesions retard context conditioning; unlike previous work that has made use of conditioned freezing as the measure of context conditioning, the present results are not explicable in terms of lesion-induced changes in general activity.

Although orthodox classical conditioning is not usually impaired in rats with hippocampal lesions, evidence has begun to accumulate in recent years to suggest that an intact hippocampus is necessary for rats to show normal conditioning to contextual cues. Initial indications came from experiments using a place-preference measure (e.g., Selden, Everitt, Jarrard, & Robbins, 1991; but see also Winocur, Rawlins, & Gray, 1987), but the bulk of recent evidence comes from experiments that measure the extent to which rats show freezing in an experimental chamber in which shocks have occurred (e.g., Kim & Fanselow, 1992; Kim, Rison, & Fanselow, 1993; Maren & Fanselow, 1997; Phillips & LeDoux, 1992, 1994, 1995). Thus, for example, Phillips and LeDoux (1992) gave rats two sessions of training, each of which contained two presentations of a tone followed by footshock. Control subjects developed a tendency to freeze not only when the tone was sounded but also in the absence of the tone, a result taken to indicate the acquisition of associative strength by contextual cues. But rats with electrolytic lesions of the dorsal hippocampus, although they showed normal acquisition of freezing in the presence of the tone, continued to remain active in its absence. They apparently had failed to form a context–shock association. One interpretation of this dissociation is that hippocampal involvement is required when the event to be learned about is complex and multifaceted, as a context may be assumed to be.

In the experiments reported in this article, we examine contextual conditioning using a different training preparation, one in which exposure to a distinctive context is accompanied by illness produced by an injection of lithium chloride (LiCl). In intact rats, this procedure generates an aversion to the context, which has been interpreted as reflecting the formation of a classically conditioned association, with the complex of cues that constitute the context as the conditioned stimulus (CS) and the nausea induced by the injection as the unconditioned stimulus (US). (See, e.g., Best, Brown, & Sowell, 1984; Symonds & Hall, 1997.) The question of interest is how lesions of the hippocampus will influence the acquisition of this association.

One reason for adopting this training procedure is that to find a lesion-induced deficit here will extend the generality of previously reported effects. Most previous studies have used footshock as the US, and there is some evidence that contextual conditioning can proceed normally in hippocampal subjects when food is used as the US (Good & Honey, 1991, Experiment 3). The use of a different US (although, admittedly, still an aversive one) allows the possibility of demonstrating that the context-conditioning deficit in hippocampal rats is not restricted to cases in which shock is used. A second and more fundamental reason is that this procedure allows us to assess the validity of an alternative interpretation of the results generated by experiments that use freezing as their measure of context conditioning.

There is a scattered but extensive literature indicating that rats with hippocampal lesions show an increase in general activity (e.g., Douglas & Isaacson, 1964; Roberts, Dember, & Brodwick, 1962; Teitelbaum & Milner, 1963), perhaps because they continue to emit exploratory responses when normal subjects have shown habituation (see Macphail, 1993). Such hyperactivity, of course, competes with any tendency to freeze in a given environment; Blanchard,

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Blanchard, Lee, and Fukunaga (1977) noted that rats with hippocampal lesions showed reduced freezing in the open field because they appeared to be poor at remaining immobile: These rats initiated freezing bouts as did control subjects, but each bout was of shorter duration. Similar effects can be expected to operate in studies of contextual conditioning, leading some to propose (e.g., McNish, Gewirtz, & Davis, 1997) that hippocampal subjects may form a perfectly normal context–US association but fail to show it when the response measure is freezing.[1] Evidence in favor of this proposal comes from experiments by Good and Honey (1997), Maren and Fanselow (1997), and by Maren, Aharonov, and Fanselow (1997). In these studies, it was found that lesions that resulted in a deficit in the acquisition of conditioned freezing to the context also produced enhanced levels of activity in the training context even before the delivery of the first shock. Although they considered other alternatives, all of these authors accepted the possibility that the freezing deficits they observed were a direct consequence of the lesion-induced occurrence of a competing response (locomotor activity). Furthermore, Good and Honey (1997) went on to offer a number of possible reasons why this heightened activity might fail to be apparent in the presence of an explicit CS (see also McNish et al., 1997).

An argument against the competing-response account of the hippocampal freezing deficit comes from the observation, made by Maren and Fanselow (1997), that there is no within-group correlation between an individual's score on the activity measure and its score on the freezing measure. This argument cannot be decisive, however. Although the presence of such a correlation would lend support to the account, the absence of the correlation does not disprove it, because there are several possible reasons why the correlation might fail to hold. For example, performance on the freezing measure is influenced by two factors: conditioned fear and general activity. If, as seems likely, there are individual differences in the readiness with which rats acquire the former, then performance on the freezing test will fail to correlate with scores obtained from a pure measure of general activity. It could nonetheless still be true that the overall difference between hippocampal and control groups in their performance on the freezing test is a consequence of enhanced general activity in the hippocampals.

We conclude that there is, as yet, no evidence to rule out the hypothesis that the freezing deficit shown by hippocampal rats is a direct consequence of the hyperactivity these lesions produce. It is thus of theoretical importance to conduct an assessment of the effects of hippocampal lesions on contextual conditioning using a response measure less likely than freezing to be distorted by changes in the rats' general level of activity. This is the central purpose of the experiments reported here.

Experiment 1

This experiment used two groups of rats: sham-operated controls (sham group) and rats with lesions of the dorsal hippocampus (hip group). The lesions were made electrolytically, this being the procedure used in the bulk of previous studies of contextual fear conditioning. (Indeed, there is reason to think that effects on context conditioning may depend on the use of electrolytic lesioning techniques. Maren et al., 1997, found neurotoxic lesions to be without effect [but see also Good & Honey, 1997].) All rats received an initial phase of training, in which they were given exposure to a distinctive context. For half the rats in each group (hip–li and sham–li groups) experience of the context was accompanied by an injection of LiCl. For the remaining subjects (hip–sal and sham–sal groups), the injection was of isotonic saline. We assessed the strength of the aversive properties acquired by the context, in a subsequent test in which the rats were given access to a saccharin solution in the training context. Many experiments conducted with unoperated animals showed that rats given context–illness pairings were less willing than controls to drink a novel-flavored solution in the trained context (e.g., Best et al., 1984; Best, Batson, Meachum, Brown, & Ringer, 1985; Boakes, Westbrook, & Barnes, 1992); thus, the degree to which ingestion was suppressed in the presence of pretrained contextual cues was widely used as measure of context conditioning. Sham–li subjects could, therefore, be expected to consume less of the saccharin than sham–sal subjects; but if hippocampal lesions interfered with context conditioning, the difference between hip–li and hip–sal groups would be abolished or attenuated. It was not obvious that lesion-induced changes in general activity could easily explain such a pattern of results.

Method

Subjects and surgery. The subjects were 32 male hooded Lister rats, approximately 3 months old at the start of the experiment. They were assigned at random to one of two equal-sized groups: hip or sham. For surgery, the rats were anesthetized with an intraperitoneal injection of Avertin at a dose of 10 ml/kg. The top of the rat's head was shaved and washed with an antiseptic solution. The rat was then placed in the ear bars of a stereotaxic frame. An incision was made, and the skin and tissue were reflected from the skull. Two small holes were made by means of a dental drill above the intended lesion sites. Bilateral dorsal hippocampal lesions were made by passing a 2.5-mA current from a constant-current lesion maker for 25 s through a wire electrode insulated to within 0.5 mm of its tip. The lesion coordinates were 3 mm posterior to bregma, 2.5 mm L to the midline, and 3.5 mm V to the brain surface. For sham-operated subjects, the procedure was the same except that no current was passed through the electrode.

After recovery from the immediate effects of surgery, the rats were deprived of food and used in a study of appetitive conditioning with a visual cue as the CS. At the completion of this study, the rats were returned to ad-lib food and left undisturbed for 2 weeks before the present experiment was begun. At this point, they were approximately 9 months old. Such evidence as we had (albeit from an investigation of much shorter time periods) suggested that the effect of hippocampal lesions on context conditioning would be unaffected by a delay between surgery and testing (Young, Bohenek, & Fanselow, 1994).

[1] Note that measures using a place-preference test also will be sensitive to effects produced by changes in general activity.

Apparatus. The rats were housed in pairs with free access to food and water, except where indicated otherwise. Their home cages measured 35 × 22 × 19 cm and were made of opaque white plastic with a lid of wire mesh that held food and (when available) a water bottle. A layer of wood shavings covered the floor. These cages were kept in the main colony room, which was brightly lit from 0800 to 2000 hr each day. A second set of cages located in a separate room in the laboratory served as the experimental context. This room was dark, and a speaker supplied a constant background white noise with an intensity of 75 dB close to the cages. The cages were made of transparent plastic, were somewhat smaller than the home cages (20 × 20 × 32 cm), and each housed a single rat. The floor was covered with commercially obtained cat litter. Fluid was presented in these cages through inverted 50-ml centrifuge tubes equipped with stainless steel, ball bearing tipped spouts. We measured consumption by weighing to the nearest 0.5 ml. The fluids used were tap water and a 0.1% saccharin solution (1 g sodium saccharin/1 l water). The US for the conditioning trials was an intraperitoneal injection of 0.3 M LiCl at 10 ml/kg of body weight.

Procedure. Before the start of conditioning, we introduced the rats, over 3 days, to a schedule of water deprivation, with the water bottles being made available for two 30-min periods each day at 1200 hr and 1800 hr. We maintained the afternoon drinking period in the home cage throughout the experiment. On the first day of the conditioning phase, the rats were transferred to the experimental context at 1200 hr and were allowed access to water for 30 min. They were then removed, given an injection of LiCl or of isotonic saline (at 10 ml/kg) according to their group assignment, and returned immediately to the experimental context, where they remained for a further 60 min before being returned to their home cages. The next day was a recovery day, with two 30-min periods of access to water in the home cage. This 2-day cycle was repeated twice.

The test session followed the final recovery day. On this session, all rats were given access to 10 ml of the saccharin solution in the experimental context for 30 min.

Histological procedures. After behavioral testing had been completed, the brains of all of the rats in the hip group and of 4 randomly chosen rats in the sham group were prepared for histological examination. The rats were killed, the brains were removed and frozen, and 30-μm coronal sections were cut. Sections were retained at 150 μm intervals throughout the lesioned area. They were mounted and stained with cresyl violet. We inspected the sections from each rat under a binocular microscope and transferred representations of lesion-induced damage onto plates adapted from the atlas of Paxinos and Watson (1986), showing positions 2.12, 2.56, 3.14, 3.60, 4.16, and 4.80 mm posterior to bregma.

Results and Discussion

Histology. Two of the operated on subjects (1 in the hip–sal group and 1 in the hip–li group) failed to sustain appropriate damage to the hippocampus; these rats were excluded from further consideration. The remaining rats all suffered some degree of bilateral damage to the dorsal hippocampus in the absence of damage to more ventral structures. In several rats, there was evidence of damage to overlying cortex, but there was minimal damage to other structures. Figure 1 contains coronal sections through the rat brain on which are superimposed reconstructions representative of the lesion damage sustained by the rats in the hip group. For each section, the hatched area shows the extent of the lesion in the rat with the greatest amount of damage, and the solid area shows the extent of the lesion in the rat with the smallest amount of damage. There was no evidence of damage to the brain in any of the sham-operated rats that were examined.

Behavior. Figure 2 contains group mean scores for water consumption during the conditioning phase. An error on the part of the experimenter meant that data were lost for 1 rat in the sham–sal group. This, with the exclusion of rats showing inappropriate lesion damage (see above), meant that the scores shown in the figure were based on a group size of 7 for each group except the sham–li group, where $n = 8$. The figure shows that the control groups given saline injections maintained their level of consumption but that consumption declined for the subjects given LiCl injections. There was no apparent difference between lesioned and sham-operated rats. An analysis of variance (ANOVA) was conducted on the data summarized in the figure, with lesioned status (hip or sham), injection type (li or sal), and trial as the variables. (In this and all subsequent statistical analyses, a criterion of significance of $p < .05$ was adopted.) There was no significant main effect of lesion ($F < 1$), but there were significant effects of trial, $F(2, 50) = 24.49$, and of the type of injection, $F(1, 25) = 15.01$. The interaction between trial and injection type also was significant. No other interactions achieved significance, Fs < 2 in all cases apart from the interaction between lesion and trial, where $F(2, 50) = 2.39$. The interaction between injection type and trial was explored by means of an analysis of simple effects, which showed the li rats differed significantly from the sal rats on Trial 3, $F(1, 73) = 39.87$.

The decline in consumption shown by the hip–li and sham–li groups might be taken to be a consequence of the formation of a context aversion and thus to imply that these groups did not differ in their ability to show contextual conditioning. But this conclusion would be premature, because the effect seen in these groups might be a consequence not of context conditioning, but of the formation of an aversion to water itself (see, e.g., Revusky & Parker, 1976, for evidence that such an aversion can be formed). The similarity in performance between the hip–li and sham–li groups might thus constitute only a further demonstration that simple conditioning could proceed normally in animals with hippocampal lesions.

This is not to assert that the context plays no role in the performance seen in Figure 2; after all, the rats that rejected water in the context were observed to drink water perfectly readily in their home cages. But again, this fact does not necessarily speak to the issue of context aversion conditioning. It has been shown that control by contextual cues does not depend on a direct association between the context and the US (Bonardi, Honey, & Hall, 1990); rather, the context appears to act as an occasion setter that facilitates the expression of an aversion formed in its presence (see also Boakes, Westbrook, Elliott, & Swinbourne, 1997; Puente, Cannon, Best, & Carrell, 1988). The results of the conditioning phase of this experiment are thus consistent with the interpretation that the acquisition of conditional contextual control over a simple association proceeds normally in rats

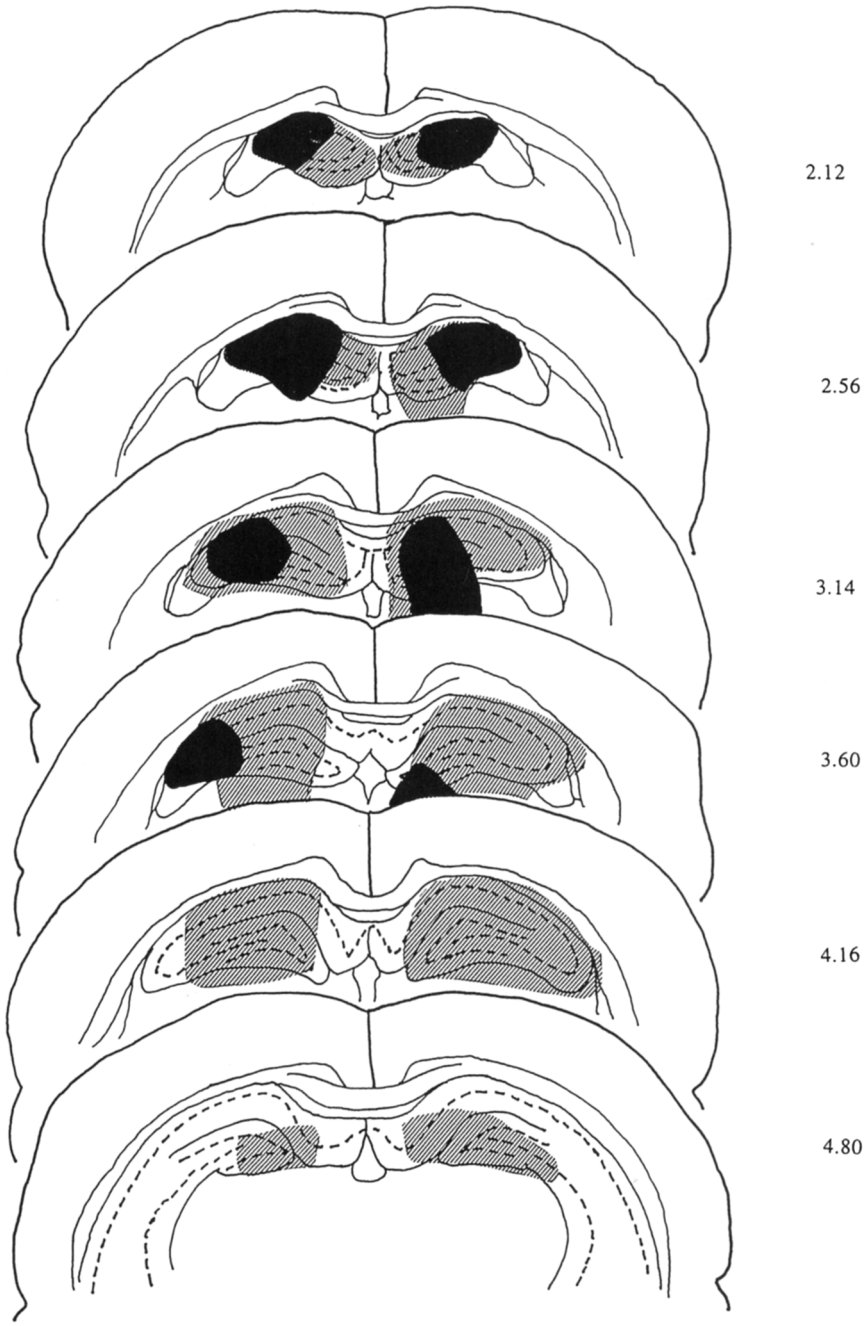

Figure 1. Experiment 1: Reconstructions of the maximum (hatched) and minimum (solid) extent of damage in the hippocampal lesioned group, superimposed on coronal sections derived from the Paxinos and Watson (1986) atlas. Figures denote positions posterior to bregma.

with hippocampal lesions. This conclusion is in accord with the results of recent experiments that have investigated this issue directly (Hall, Purves, & Bonardi, 1996; Wilson, Brooks, & Bouton, 1995; but see also Good & Honey, 1991; Honey & Good, 1993).

Better evidence on the strength of the aversive properties acquired by the context comes from the results of the saccharin consumption test. Group mean scores are presented in Figure 3. This shows that the groups given saline injections in the training phase drank more than the groups given LiCl injections. Hippocampal lesions had no effect on the performance of the saline-injected rats, but the hip–li group showed less suppression of consumption than the sham–li group. ANOVA was conducted on these data, with lesion (hip or sham) and injection type (li or sal) as the variables. This yielded significant effects of lesion, $F(1, 25) = 4.69$, and of injection-type, $F(1, 25) = 113.77$, and a significant interaction between these variables, $F(1, 25) =$

4.26. Pairwise comparisons among individual group means using Tukey's test showed that each group differed significantly ($p < .05$) from each of the others, apart from the hip–sal and sham–sal group, which did not differ from each other. To the extent that suppression of saccharin consumption is an accurate measure of the aversive properties acquired by the context, we may conclude that such properties were acquired in the groups given lithium injections but that acquisition of the context aversion was retarded in the hippocampal subjects. These results are thus consistent with the conclusions derived from studies that have used shock as the US and freezing as the response measure: An intact hippocampus is necessary for normal context conditioning.

Note, however, that the results presented in Figure 3 may be open to other explanations. Three are discussed, all of which take issue with the assumption that suppression of saccharin consumption shown by the li groups is an accurate measure of the strength of a context–illness association. First, it is possible that the performance of the li groups reflects not a context aversion, but generalization to saccharin of the aversion acquired to water during the conditioning phase (see Symonds & Hall, 1997). The low level of saccharin consumption shown by the hip–li group would thus indicate enhanced acquisition of the original water aversion rather than retarded acquisition of the context aversion. This possibility seems unlikely, however, given the fact that the hip–li and sham–li groups showed no sign of a difference in their water consumption during conditioning. In this respect, the present results match those of earlier experiments that have found flavor aversion learning to be unaffected by hippocampal damage (e.g., Murphy & Brown, 1974; Purves, Bonardi, & Hall, 1995).

The second possibility to be considered is that the test

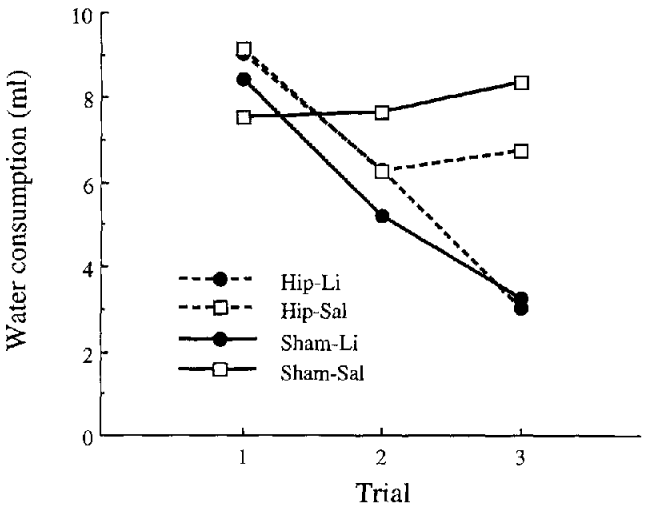

Figure 2. Experiment 1: Group mean consumption of water (in milliliters) during the context-conditioning phase. All four groups were given access to water in the context. Li groups received an injection of lithium chloride after being taken out of the context; sal groups received an injection of saline. Hip groups had sustained hippocampal lesions; sham groups underwent a sham operation.

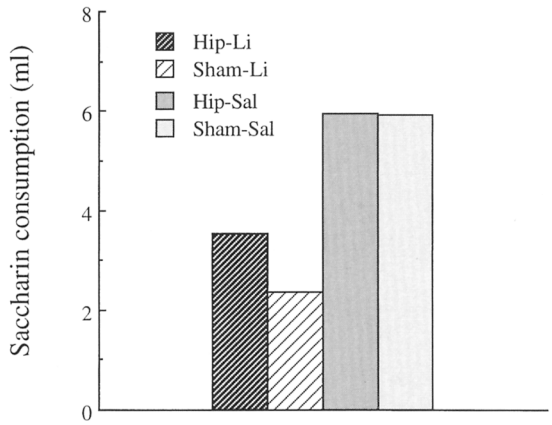

Figure 3. Experiment 1: Group mean consumption of saccharin (in milliliters) on the test trial in the context that in the previous stage had been associated with an injection of lithium chloride for the li groups and a saline injection for the sal groups. Hip groups had sustained hippocampal lesions; sham groups underwent a sham operation.

results of this experiment are determined in part by the rats' neophobic response to saccharin. The suppression of consumption shown by the sham–li group could be a combination of context aversion plus neophobia. Now previous studies of the response to novel flavors of rats with hippocampal lesions have demonstrated that neophobia is often reduced (Kranc, Sinnamon, & Thomas, 1976; Miller, Nonneman, Kelly, Niesewander, & Isaac, 1986). Thus, context conditioning might proceed quite normally in the hip–li group, but if their initial response to novelty was weak, the neophobic component of response suppression would be absent for these rats, and they would drink more saccharin on test than the sham–li group. A reason to question the validity of this interpretation comes from the fact that the hip–sal and sham–sal groups did not differ in their test performance; an effect of hippocampal lesions on neophobia might be expected to show in these groups as in those given context conditioning. It is necessary to assume, therefore, that the effect of hippocampal damage on neophobia will be evident only in animals that have received prior experience of poisoning. This is clearly possible, but there is little evidence to support the assumption: Those studies that have obtained an attenuation of neophobia in rats with hippocampal lesions have done so for rats that had no prior experience of poisoning.

Finally, note the possibility that the amount of saccharin consumed on the test might conceivably be influenced by lesion-induced changes in activity. Although the response measure used here seems less likely than freezing to be sensitive to such effects, it may not be wholly immune. If hippocampal animals are hyperactive because they persist in exploring their environment when normal animals have stopped (see Good & Honey, 1997), then engaging in such exploration could increase the likelihood that the lesioned animals might rapidly encounter the drinking spout, and this might be responsible for the higher level of saccharin intake in the hip–li group. As it stands, this hypothesis is unable to

explain why the hip and sham groups did not differ in their water consumption during training; nor can it explain why the hip–sal and sham–sal groups failed to differ on their saccharin test. Nonetheless, it seems worthwhile to address the matter directly by carrying out a further study of context aversion learning, making use of a test procedure in which animals that fail to show normal context conditioning can be expected to drink less, not more, of the test fluid. Such a test would also allow us to provide evidence for an effect of hippocampal lesions on context conditioning that could not be explained in terms of a lesion-induced attenuation of neophobia.

Experiment 2

Symonds and Hall (1997) made use of a blocking procedure to assess the strength of context aversion learning in intact rats. In their Experiment 2, the rats experienced two distinctive contexts in the first stage of training; one (Context A) was paired with an injection of LiCl, and one (Context B) was not. In the next stage of training, the rats consumed sucrose in their home cages and were then placed in one or the other of the contexts before receiving a further injection of LiCl. For rats that experienced Context B in this stage, this serial compound conditioning resulted in a strong aversion to sucrose (tested in the home cage); animals that experienced A in this stage consumed sucrose more readily. Symonds and Hall concluded that an aversion formed to Context A in the first stage of training had blocked the acquisition of the aversion to sucrose.

The present experiment included a pair of groups (sham–A and sham–B, where *A* and *B* referred to the context presented in the second stage of conditioning) that exactly matched those in the experiment by Symonds and Hall (1997). A second pair of groups (hip–A and hip–B) received equivalent training but had suffered lesions of the hippocampus. We anticipated that the sham groups would replicate the effect observed by Symonds and Hall, but if hippocampal lesions prevent contextual conditioning, no blocking effect would be evident in the hip groups; that is, the hip–A group would consume sucrose on the test no more readily than the hip–B group. Note that with this training procedure, a failure of context conditioning would be revealed by a low level of consumption on the final test. Enhanced consumption as a consequence of enhanced exploratory activity in hippocampal subjects could thus not be responsible for such an outcome.

Method

Subjects. The subjects were 32 male hooded Lister rats, about 3 months old at the time of surgery. Sixteen received bilateral hippocampal lesions; the procedures used were the same as those described for Experiment 1. The remaining 16 underwent a sham operation. After they had recovered from surgery, they were deprived of food and used in a study of appetitive Pavlovian conditioning with food as the US and light and noise as the CSs. At the end of this study, they were returned to ad-lib food and water in preparation for the present experiment, which began when the rats were approximately 7 months old. On completion of this experiment, the rats were killed and their brains prepared for histological examination, as described in Experiment 1.

Apparatus. The apparatus was that used in Experiment 1, with the addition of a further set of cages that constituted a second experimental context. These were larger than the home cages, measuring 42 × 35 × 16 cm, and were located in a brightly lit colony room that contained racks of standard cages housing other rats. The walls and floors of the cages were made of translucent white plastic, and the wire mesh roofs included a section through which a drinking spout could be inserted. The floors were left bare. Previous work (e.g., Symonds & Hall, 1997) had shown that these cages were discriminably different from the smaller cages used as the experimental context in Experiment 1.

Procedure. A schedule of water deprivation was established as in the previous experiment. The next 8 days constituted the context-conditioning phase. On Day 1, all subjects were put into Context A for 30 min at 1200 hr and given access to 10 ml of water. They were then removed from the context and immediately given an injection of LiCl before being returned to their home cages. As in the previous experiment, the subjects were allowed, in the home cage, 30 min of free access to water in the standard bottles later in the day (at 1700 hr). On Day 2, the subjects were placed in Context B at 1200 hr, given 10 ml of water for 30 min, and then returned to the standard home cages, no injection having been given. Again, the subjects were given free access to water in the home cage at 1700 hr. This 2-day cycle was then repeated a further three times. For half the subjects in each group (hip and sham), the large cages served as the conditioning context (A) and the smaller cages as the no-injection context (B); for the remainder, this arrangement was reversed.

Following the procedure used by Symonds and Hall (1997), there was a 6-day interval between context conditioning and the next phase of training, during which the rats were given access to water, twice daily, in their home cages. All animals then received a single compound-conditioning trial. At 1200, they received a 10-ml presentation of a 0.33 M solution of sucrose for 15 min in the home cage. They were then transferred to one of the experimental contexts for 30 min, removed, and immediately injected with LiCl before being returned to the standard housing racks. For half the subjects in each group (hip–A and sham–A), the context used on this trial was the context that had previously been paired with illness. For the remaining subjects (the hip–B and sham–B groups), this context was the no-injection context. On the next day, subjects were given a recovery day on which they received two 30-min sessions of free access to water in the home cage, at 1200 hr and 1700 hr. Finally, there was a nonreinforced test in which free access to sucrose was given in the home cage for 15 min at 1200 hr. Any procedural details not specified here were the same as those described for Experiment 1.

Results

Histology. One of the rats in group hip–A failed to sustain damage to the hippocampus and was excluded from the behavioral analysis. For 2 further rats (1 in group hip–A and 1 in group sham–A), an error on the part of the experimenter resulted in their brains not being available for histological examination. Accordingly, these 2 rats also were excluded from further analysis, resulting in final group sizes as follows: sham–A, 7; sham–B, 8; hip–A, 6; group hip–B, 8. All of the remaining rats in the hip group sustained bilateral damage to the dorsal hippocampus; there was some damage to overlying cortex but little sign of damage to other structures. Figure 4 contains coronal sections through the rat

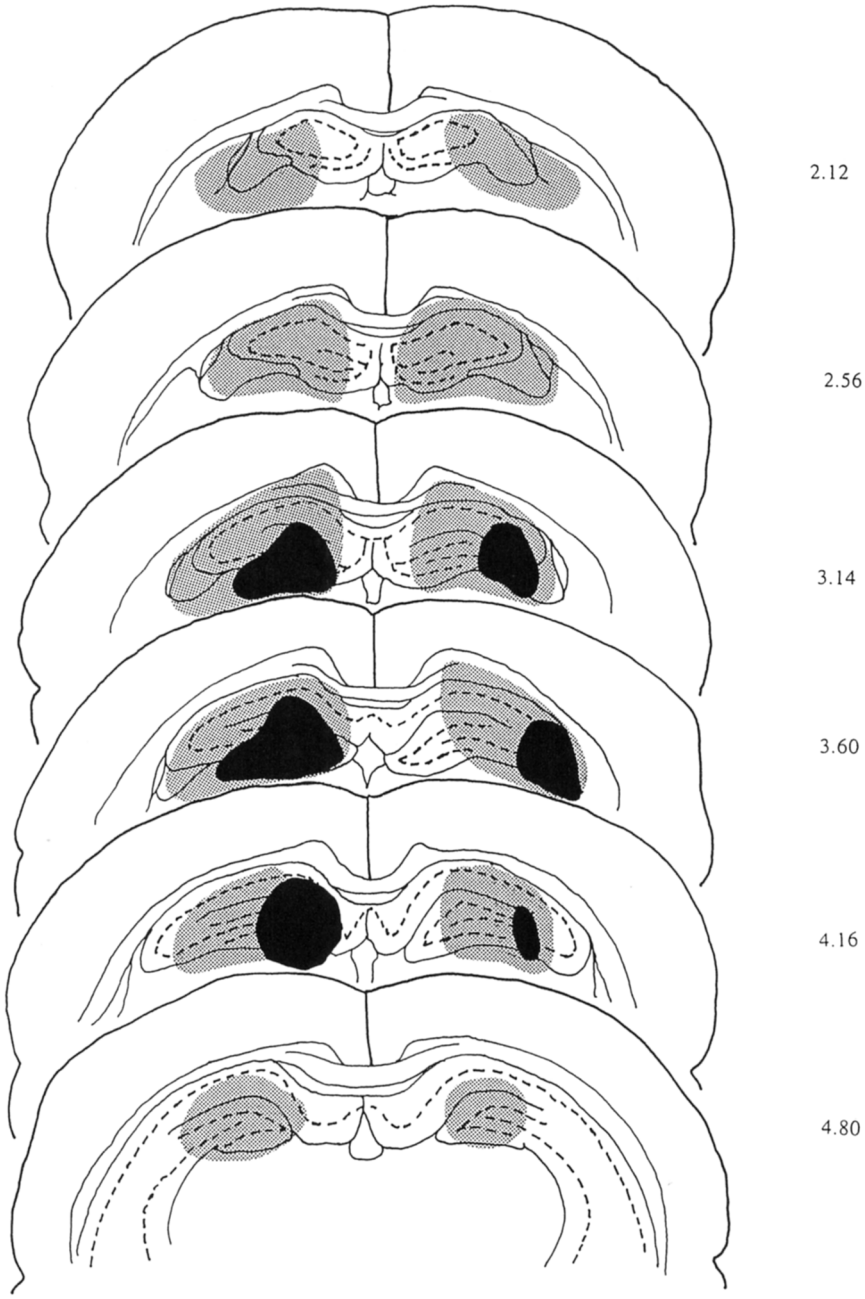

Figure 4. Experiment 2: Reconstructions of the maximum (stippled) and minimum (solid) extent of damage in the hippocampal lesioned group, superimposed on coronal sections derived from the Paxinos and Watson (1986) atlas. Figures denote positions posterior to bregma.

brain on which are superimposed reconstructions representative of the lesion damage sustained by the animals in the hip group. For each section, the stippled area shows the extent of the lesion in the rat with the greatest amount of damage, and the solid area shows the extent of the lesion in the rat with the smallest amount of damage. There was no evidence of damage to the brain in any of the sham-operated rats.

Behavior. Figure 5 shows the mean amount of water consumed by the hip rats and the sham rats in each context, during the first phase of training. It is evident that this training schedule, in which consumption of water was followed by an injection of LiCl only on alternate days, was less effective in suppressing consumption than the reinforcement schedule used in Experiment 1. Nonetheless, by the final trials of this phase, a discrimination had developed, with the rats drinking less in the lithium chloride associated Context A than in Context B. This was true both for sham and hip groups. We conducted an ANOVA on the data summarized in the figure, with trial (1–4), context (A or B), and group (hip or sham) as the variables. There was a

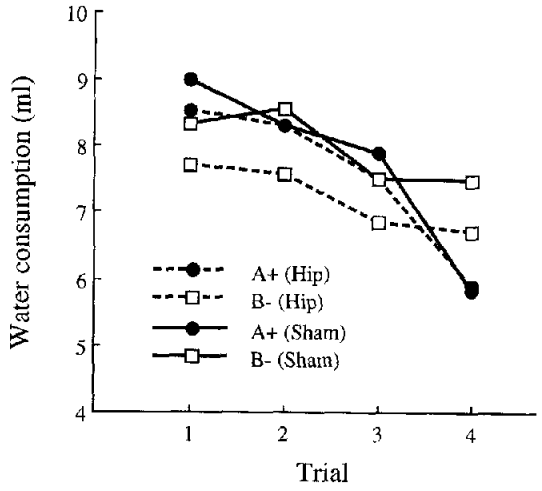

Figure 5. Experiment 2: Group mean amounts of water consumed (in milliliters) during context conditioning. All subjects experienced two contexts, one (A) associated with an injection of lithium chloride (+) and the other (B) not associated with an injection (−). Rats in the hip groups had sustained hippocampal lesions; rats in the sham groups underwent a sham operation.

significant main effect of trial, $F(3, 81) = 18.29$, but not of group, $F(1, 27) = 1.45$, or of context ($F < 1$). None of the interactions was significant (Fs < 1) except for that between trial and context, where $F(3, 81) = 7.94$. An analysis of simple main effects showed that on Trial 4, consumption in Context A differed reliably from consumption in Context B, $F(1, 27) = 9.65$.

On the compound-conditioning trial, all subjects drank the sucrose readily. The mean scores were 7.92 ml for the Hip–A group, 8.56 ml for the hip–B group, 8.29 ml for the sham–A group, and 8.63 ml for the sham–B group. An ANOVA with group and context as the variables yielded no significant effects; all Fs < 1, apart from that for the A versus B variable, where $F(1, 25) = 1.79$.

The data of central interest are those for sucrose consumption on the test trial. Group means are presented in Figure 6. The results for the sham groups replicate those reported by Symonds and Hall (1997) in that those rats that experienced the previously reinforced context (sham–A group) on the compound-conditioning trial consumed the sucrose readily, whereas those that experienced the nonreinforced context (sham–B group) on this trial consumed rather less. This is consistent with the interpretation that for the sham rats, Context A was capable of blocking the acquisition of an aversion to sucrose and, thus, that a Context A–illness association was formed in these rats during the first stage of training. The hippocampal groups, by contrast, did not differ in the amount of sucrose they consumed on the test trial, implying that context conditioning was disrupted in some way for these rats. We conducted an ANOVA on the data shown in Figure 6, with lesioned status (sham or hip) and compound-conditioning context (A or B) as the variables. There was no main effect of lesion ($F < 1$) or of context, $F(1, 25) = 2.87$, but there was a significant interaction between these variables, $F(1, 25) = 5.54$. This interaction was explored by means of an analysis of simple effects, which revealed a significant effect of context type in the sham groups, $F(1, 25) = 8.56$, but not in the hippocampal groups ($F < 1$). Pairwise comparisons using Tukey's test confirmed that the sham–A group differed significantly ($p < .05$) from the sham–B group but revealed no other significant effects.

Discussion so far has focused on the fact that the hip–A and hip–B groups did not differ on the test. But the absolute level of consumption shown by these groups requires comment. This level was intermediate between that shown by the two sham groups. On the face of things, this is not what would be expected if the sole effect of hippocampal lesions in this training procedure was to prevent context conditioning: A simple abolition of context conditioning would mean that each of the hip groups would develop an aversion to sucrose comparable to that shown by the sham–B group. One possible explanation for the elevated level of consumption shown by the hip groups follows from the observation (noted above) that rats with hippocampal lesion may show a reduction in neophobia. Because the test trial in this experiment constitutes only the second occasion on which the rats have experienced sucrose, it is possible that the suppression of consumption shown by the sham–B group reflects a combination of the aversion acquired on the previous trial and a neophobic response. The hip groups, lacking the full neophobic response, can thus be expected to consume more on this test.

A second possible explanation for the pattern of results shown in Figure 6 makes use of the notion of *superconditioning*. Just as a pretrained excitatory CS can block the acquisition of associative strength by another stimulus with which it is reinforced in compound, so a pretrained inhibi-

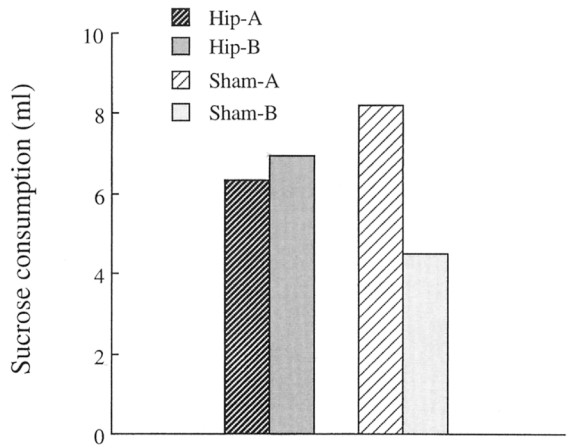

Figure 6. Experiment 2: Group mean consumption of a sucrose solution (in milliliters) presented in the home cage on the final (nonreinforced) test trial. Hip–A and sham–A groups previously had experienced sucrose followed by exposure to Context A and an injection of lithium chloride (LiCl); hip–B and sham–B groups previously had experienced sucrose followed by exposure to Context B and an injection of LiCl. Rats in the hip groups had sustained hippocampal lesions; rats in the sham groups underwent a sham operation.

tory CS might be able to enhance the conditioning occurring to the other element of the compound (Rescorla, 1971). For the sham groups of the present experiment, the context-training procedure might well be capable of establishing not only Context A as an excitor but also Context B as an inhibitor for the US of the lithium chloride injection. If so, the test performance of these groups would be a consequence both of blocking of the sucrose aversion in the sham–A group and of superconditioning of this aversion in the sham–B group. If hippocampal subjects were incapable of learning about contexts, they would show neither of these effects, and the result would be an intermediate level of sucrose consumption.

Finally, note that the use of the blocking procedure in this experiment introduces an additional complication. It has been suggested by a number of authors (e.g., Gallo & Cándido, 1995; Rickert, Bennett, Lane, & French, 1978) that hippocampal lesions can attenuate or abolish the blocking effect. If so, then the results of the present experiment might reflect not a failure of contextual conditioning in hippocampal subjects, but a failure of the context–illness association to exert a blocking effect on the acquisition of the sucrose aversion. There is nothing in the data reported here that allows us to argue either for or against this interpretation, but two points may be made. First, the notion that hippocampal lesions abolish blocking has been a matter for debate, and studies have been conducted that reveal no sign of such an effect, even in rats that have suffered very substantial hippocampal damage (e.g., Garrud et al., 1984). Second, the results of our experiment accord well with those of Experiment 1, in which the blocking procedure was not used. The proposal that hippocampal lesions interfere with the rat's ability to form context–illness associations thus provides a parsimonious account of the entire set of results.

General Discussion

The aim of the experiments reported here was to examine the effects of lesions of the dorsal hippocampus on conditioning when contextual cues constituted the CS and illness produced by an injection of LiCl constituted the US. Previous results purporting to demonstrate a deficit in context conditioning had come from studies of fear conditioning that used footshock as the US and measured freezing as the conditioned response.

Interpretation of these studies is made difficult by the fact that hippocampal lesions have been shown to result in hyperactivity: A low level of freezing in hippocampal subjects can be a direct effect of this hyperactivity rather than an inability to learn about the context. The procedure used to assess context conditioning in the present experiments produces effects that are resistant to explanation in terms of lesion-induced changes in general activity. In both our experiments, the behavior measured on test was the amount of a fluid consumed. In Experiment 1, however, the critical result was the demonstration that rats with hippocampal lesions drank more than control subjects, whereas in Experiment 2, the critical finding was that they drank less than did rats in one of the control conditions. It is difficult to see how an enhanced level of general activity could generate both these results.

Eliminating this possible artifact prompts further consideration of the proposal that lesions of the hippocampus selectively disrupt conditioning of contextual cues. Why this might be so is still a matter for speculation, but one suggestion is that to function effectively as a CS, the complex of cues that form a context must undergo some form of processing that assembles the elements of the complex into a unified or integrated stimulus representation (e.g., Fanselow, 1990; Young et al., 1994; see also Good & Honey, 1997). The mechanism responsible for this processing is assumed to require an intact hippocampus. Although the evidence on which this proposal is based comes from experiments involving shock as the US, an effect of hippocampal lesions should be found in any procedure in which subjects are required to learn about a context. Our results provide only mixed support for this proposal. To the extent that they show a deficit in the formation of a context–illness association, they may be taken to support this account. On the other hand, our experiments (along with others, e.g., Hall et al., 1996; Wilson et al., 1995) have failed to demonstrate any deficit in the acquisition of a discrimination in which the context served as a conditional cue, indicating whether some other event (consumption of water in these experiments) would be followed by the US. It is difficult to see why the processing that is needed for a context to function as a simple CS would not also be needed for the context to function as a conditional cue; current theories will need elaboration if they are to deal successfully with this issue.

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