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Pharmacology of Sucrose-Reinforced Place-Preference Conditioning: Effects of Naltrexone

ANDREW R. DELAMATER,* ANTHONY SCLAFANI,* AND RICHARD J. BODNAR†

*Experimental Psychology, Brooklyn College, Brooklyn, NY; and †Neuropsychology Doctoral Subprograms, Queens College, City University of New York, New York, NY

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DELAMATER, A. R., A. SCLAFANI AND R. J. BODNAR. *Pharmacology of sucrose-reinforced place-preference conditioning: Effects of naltrexone.* PHARMACOL-BIOCHEM BEHAV **65**(4) 697–704, 2000.—Two experiments investigated the role of the opioid system in sucrose-reinforced conditioned place preferences (CPPs) in rats. Experiment 1 examined the effects of a general opioid antagonist, naltrexone, on the expression of a CPP acquired in the absence of the drug. Subjects were trained to associate one compartment of a two-compartment chamber with sucrose and the other compartment with water. Rats displayed a preference for the sucrose-associated compartment in a choice test without sugar or water available following vehicle saline treatment. Naltrexone doses of 2.5 and 5.0 mg/kg reduced this preference for the sucrose-associated compartment. Experiment 2 examined the effects of naltrexone on the acquisition as well as the expression of CPPS. Different groups of rats received daily injections of either saline, 0.1, 1.0, or 5.0 mg/kg of naltrexone prior to each training session, and then these groups were given a choice test for the CPP after saline or naltrexone injections. Although naltrexone treatment attenuated the expression of CPPs in each group relative to saline treatment, there were no group differences during these tests in the magnitude of the preferences. Moreover, all groups displayed equal acquisition of CPPs despite the fact that naltrexone dose dependently decreased sucrose intake during the training phase. Together, the results indicate that the opioid system modulates the expression but not the acquisition of sucrose-reinforced CPPs. © 2000 Elsevier Science Inc.

Conditioned place preference Sucrose Naltrexone Opioids Acquisition Expression

THE conditioned place preference (CPP) procedure has been used extensively to investigate the neuropharmacological substrates of reinforcement (34,35). In a typical procedure, the rat is taught to associate one compartment of a two-compartment chamber with some reinforcing agent, and the other compartment with the absence of that reinforcing agent. Following such training, the animal is given a choice between the two compartments in the absence of the reinforcer. A preference for the reinforced compartment in this test is often thought to result from a Pavlovian conditioning process, in which the reinforcer–paired compartment can be designated as the reinforced conditioned stimulus (CS+), and the alternate compartment as the nonreinforced conditioned stimulus (CS−).

The vast majority of CPP studies has focused on pharmacological effects on place preference learning established by drug reinforcers [e.g., (35,37,40)], rather than by "natural" reinforcers such as food. A growing literature, however, has demonstrated that CPPs can be established using natural reinforcers, and that opioid and dopaminergic systems may play important roles in this learning.

Place preferences in food-restricted rats have been demonstrated with sucrose solutions, sucrose–mash, sucrose pellets, and plain pellets [eg., (1,14,27,29,39,43)]. Moreover, although these studies have begun to explore the involvement of the opioid and dopaminergic systems in the learning of these place preferences, a clear picture has not emerged. For instance, to our knowledge there has only been one study examining the effects of opioid antagonists on food-reinforced CPPS, and this study provided some support for a role of the opioid system (1). Naloxone administered during training not only reduced the CPP (during a subsequent drug-free test), but moderate to high doses produced place aversions.

Requests for reprints should be addressed to Andrew R. Delamater, Psychology Department, Brooklyn College–CUNY, 2900 Bedford Ave., Brooklyn, New York 11210. email address: andrewd@brooklyn.cuny.edu

Investigations into the role of dopamine in food-reinforced CPPs have yielded mixed results. Whereas haloperidol administered during the CPP training phase reduced a food-reinforced CPP in a drug-free test (39), other studies (9,14) demonstrated that the learning of food-reinforced CPPs was facilitated by specific D_2 (sulpiride, pimozide, and amisulpride) but not by less specific or D_1 (haloperidol, metoclopramide, chlorpromazine, or SCH 23390) dopamine antagonists. In addition, Ågmo et al. (1) observed that a mixed D_1/D_2 dopamine antagonist, flupentixol, blocked a sucrose-reinforced CPP.

Interpretations of these results, however, are complicated because these studies have confounded the putative modulatory effects of the drugs upon reinforcement processes with potential direct drug effects on place conditioning per se. More specifically, these studies used a conditioning procedure in which the drug was administered only during sessions in which food was paired with the CS+ compartment. During nonfood sessions, subjects were injected with saline prior to being confined to the CS− compartment. Thus, not only could the animals learn to associate the CS+ compartment with food, and the CS− compartment with no food in these studies, but they could also learn to associate the CS+ selectively with the physiological consequences of the drug.

Spyraki and coworkers (39) recognized this potential problem and attempted to eliminate it by demonstrating in a control experiment that no CPP or conditioned place aversion occurred when the drug alone, without food, was associated with the CS+ compartment. Nevertheless, although this failure to find direct evidence for place-drug learning is suggestive, it remains possible that such learning can be more easily revealed as a competing effect upon food-reinforced place conditioning. Therefore, it is important to determine whether any reported drug effects upon food-reinforced CPPs would occur in a situation in which this potential problem has been controlled. One purpose of the present studies was to create such a situation.

A second purpose of the present studies was to explore separately drug effects upon the acquisition and expression of a food-reinforced CPP. All of the previously cited studies investigated the effects of drugs upon the acquisition, but not expression of food-reinforced CPPs. It is conceivable that drug effects on the acquisition and expression of food-reinforced CPPs are dissociable.

Given the relative paucity of research examining the involvement of the opioid system in food-reinforced CPPs together with the established effect of opioid antagonists upon sucrose-mediated behaviors [see reviews: (8,10,18)], the present studies explored the effects of an opioid antagonist, naltrexone, upon a sucrose-reinforced CPP. Experiment 1 explored the effects of naltrexone on the expression of a previously learned CPP, and Experiment 2 explored the effects of naltrexone both on the acquisition and the expression of a CPP.

EXPERIMENT 1: EFFECTS OF NALTREXONE UPON EXPRESSION OF A SUCROSE-REINFORCED CPP

Method

Subjects. Sixteen experimentally naive male Sprague–Dawley rats (Charles River Laboratories, Wilmington, MA) weighing 370–410 g at the beginning of the experiment were used. They were individually housed in a colony room that was on a 16 L:8 D cycle, and they were maintained with daily supplemental feedings at 85% of their ad lib body weights. Food rations were given approximately 2 h following an experimental session that occurred approximately 4 to 5 h into the light cycle.

Aparatus. The apparatus consisted of eight identical two-compartment conditioning chambers each of which was housed in a sound- and light-resistant shell. The conditioning chambers measured 45.2 × 20.2 × 19.2 cm (l × w × h). The floor consisted of 0.2-cm diameter stainless steel rods spaced 1.0 cm apart, and it rested on a central pivot. The rat's location was determined electronically through the activation of a switch when the animal was in one or the other compartment. Two end walls were constructed of aluminum, while the side walls and the ceiling were made from clear Plexiglas. In the center of both end walls 3.0 cm above the grid floor was a 1.0-cm diameter opening through which a metal spout could protrude approximately 2.5 cm into the chamber. A 50-ml graduated drinking tube could be attached to the outer side of the wall, and supply fluid. During conditioning sessions, intake was recorded to the nearest 0.5 ml after the rat was removed from the chamber. A 40-W light bulb was mounted near the top right portion of the rear wall of the outer shell. This light bulb remained continuously illuminated during the session. A fan attached to the outer shell provided for ventilation as well as continuous noise. The rat could be confined to one or the other compartment by placement of a removable sheet-metal barrier that extended from the grid floor to the ceiling. The two compartments were distinguished from one another in several ways. First, the side and end walls of one compartment contained alternating 1.8-cm strips of black and white tape that were vertically oriented. The other compartment contained horizontally oriented black and white strips of tape. The central barrier contained no tape. The two compartments also differed in their proximity to the outer shell's light bulb, which was positioned slightly off center so that the two compartments were not identically illuminated. Finally, the two compartments contained different textured floors. Hardware cloth (1.3 × 1.3 cm) was attached to the grid floor of one compartment. All experimental events were controlled and recorded automatically by a microcomputer and interfacing equipment located in the same room.

Procedure. The subjects were pretested on the first day of the experiment. The rats were placed in the two-compartment apparatus for a 10-min session with the barrier removed. There were no fluids available during this session, and the amount of time spent in each compartment was recorded.

Conditioning phase. Over the next 20 sessions (30 min/day) subjects received a double-alternating sequence of differential conditioning. When confined to the CS+ side, the rats had access to a 16% sucrose solution, and when confined to the CS^- side, they had access to plain water. The position (left/right) of the CS+ side as well as the specific stimuli (horizontal/vertical) that served as the CS+ were fully counterbalanced across subjects. In addition, an unbiased place preference conditioning procedure was used in the studies reported here in that there was no group preference for either CS+ or CS^- in the pretest session.

To familiarize the rats to the drug injection procedure, they were given subcutaneous (SC) injections of saline (0.9% NaCl, 1 ml/kg body weight) 15 min prior to the beginning of their final two conditioning sessions.

Test phase. On each of 2 days following the final conditioning session, place preference tests were conducted much like the pretest had been conducted earlier. The rats were given 10-min access to the preference apparatus with the barriers removed, but with no fluids or spouts present. The rats were

administered an SC injection of either saline (0.9% NACl) or a 2.5 mg/kg dose of naltrexone HCl (Sigma Chemical Co., St. Louis, MO) 15 min prior to the test session. Over the 2-day test sequence, each rat was injected with each solution with the order counterbalanced across rats.

A second 2-day sequence of preference tests was given following four additional retraining sessions. These test sessions were performed as in the first two test sessions, except that a 5.0 mg/kg dose of naltrexone was used. The order in which rats received naltrexone or saline injections in this second set of test sessions was orthogonal to the order that they received in the first set of tests.

Statistical analysis. Standard analysis of variance (ANOVA) techniques were used throughout the article to evaluate the data. A type I error rate of 0.05 was adopted. In addition, post hoc *t*-tests were performed as reported below. Three measurements were taken: intake during the conditioning phase, the percentage of total time spent on the sucrose-associated side during a test session, and the number of times during a test session that the rats crossed into the sucrose-associated compartment. Because the number of crossover responses in naltrexone and saline tests never differed in the present studies, these data are not reported.

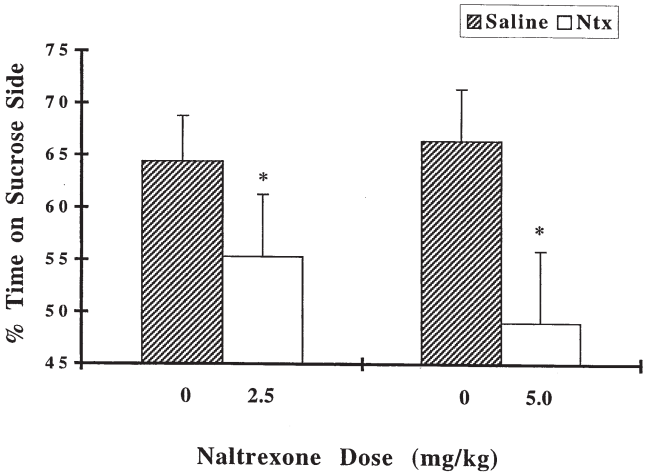

FIG. 1. Mean % time spent in the sucrose-associated compartment (+ standard error of the mean, SEM) during the test sessions of Experiment 1. Rats were tested after being injected with saline, 2.5 mg/kg naltrexone, and 5.0 mg/kg naltrexone. Asterisks denote a significant main effect comparing saline and naltrexone test sessions.

Results

Rats consumed more of the sucrose solution than plain water during the conditioning and retraining phases. Sucrose intake increased over sessions, leveling off at 25–27 ml, while water intake remained low (less than 2 ml) throughout. This difference was highly reliable in the final three 2-day cycles of training, $t(15) = 16.35, p < 0.0001$.

Figure 1 presents the place preference data across test sessions. When saline was injected prior to the test, rats displayed a 64–66% preference for the CS+ side. Naltrexone reduced this preference to 49–55%. These data were evaluated statistically with a test sessions (saline vs. 2.5 mg/kg sessions or saline vs 5.0 mg/kg sessions) × injection (saline or naltrexone) repeated-measures ANOVA. This analysis revealed that naltrexone significantly reduced the magnitude of the place preference, $F(1, 45) = 6.50, p < 0.025$. Neither the main effect of test nor the test × injection interaction were significant. Separate analyses were performed, post hoc, to determine if the percent time spent in the CS+ compartment differed reliably from 50% (i.e., a non-preference baseline). Significant increases above 50% were shown in the two saline tests, $t(15) = 3.31, p < 0.005$ and $3.27, p < 0.005$, but not in the two naltrexone tests.

Discussion

The results of the present experiment demonstrated that both doses of naltrexone reduced the expression of a previously learned sucrose-reinforced CPP. Stronger preferences for the sucrose-associated compartment were seen in the saline compared to the naltrexone tests. Moreover, preference scores failed to differ from 50% in tests with both doses of naltrexone, while significant preferences for the sucrose-associated compartment were obtained when the same subjects were tested after saline injections. Thus, these data suggest that the opioid system may be important in modulating the expression of sucrose-reinforced place preferences.

One potential problem, however, in reaching definitive conclusions about the opioid pharmacology of CPPs from these data concerns the possibility that drug novelty might have reduced the expression of the CPP during the test session. Because subjects were drug naive entering the test, the novel state produced by the drug could have nonspecifically and unconditionally disrupted the expression of the CPP.

A second potential problem in deriving conclusions from these data concern the possibility that state-dependent learning [e.g., (26)] may have influenced the test results. In other words, if the place–sucrose association (presumed to mediate the CPP) was relatively specific to the physiological state of the animal during training, then any change in physiological state produced by naltrexone may have interfered with the retrieval of that association during the test session. Experiment 2 used a design in which the contributions of novelty or state-dependent learning to CPPs could be assessed.

EXPERIMENT 2: EFFECTS OF NALTREXONE UPON THE ACQUISITION AND EXPRESSION OF A SUCROSE-REINFORCED CPP

Whereas Experiment 1 explored the effects of naltrexone only upon the expression of a previously learned CPP, the present experiment explored the effect of naltrexone upon both the acquisition as well as the expression of a CPP. Ågmo and coworkers (1) compared CPPs during a drug-free test in separate groups of rats that had either been injected with saline or naloxone during the conditioning phase. However, as noted above, the drug was injected only on CS+ sessions during the training phase in this study. Thus, this procedure permitted the possibility that any aversive properties of the drug might have directly associated with either the sucrose-associated place or with sucrose itself. Either of these possibilities could have resulted in a weaker sucrose-reinforced CPP, but for reasons other than an opioid involvement in the acquisition of food-reinforced CPPS. These possibilities are plausible in light of data indicating that naloxone had indeed conditioned a taste aversion to sucrose at doses that were also shown to convert a sucrose-reinforced CPP into a conditioned place aversion (1).

The present study attempted to avoid these problems by administering naltrexone on both CS+ and CS− days during the training of the CPP. Thus, each compartment of the CPP

apparatus is given an opportunity to associate with any potential aversive effects of the drug. Furthermore, this procedure should discourage rats from acquiring a taste aversion to sucrose based upon the putatively aversive properties of naltrexone, because although sucrose and naltrexone are paired on CS+ days, naltrexone is additionally presented in the absence of sucrose on CS− days. Separate presentations of the unconditioned stimulus (US) has been shown in numerous conditioning paradigms [including taste aversion learning; (25)] to reduce the effectiveness of CS–US pairings [e.g., (22)].

Four groups of rats participated in the present study, with three of these receiving injections of different doses of naltrexone throughout CPP training. The fourth group received saline injections throughout training. During the test phase, each group of rats was tested for their CPP following saline treatment as well as their training dose of naltrexone. Using this design, any state-dependent learning effects would be expected to result in larger CPPs in the naltrexone test than in the saline test in the three groups receiving naltrexone during training. In addition, any disruptive effects of drug novelty on the CPP should be restricted to the saline control group, as this was the only group for which naltrexone was novel in the test. Of course, it is possible that having been injected with the drug throughout training and then being tested without the drug (saline treatment) could constitute a "novel" condition. If this were the case, we would also expect greater preferences in the naltrexone test compared to the saline test in the drug-conditioned groups. In contrast, if the results of Experiment 1 were due to direct opioid involvement, then each of the groups may be expected to reveal stronger CPPs following saline relative to naltrexone during the test phase. Moreover, if naltrexone interferes with the acquisition of a CPP, then groups receiving naltrexone during training should display reductions in the size of their CPPs.

Method

Subjects. An experimentally naive group of 64 male Sprague–Dawley rats (Charles River Laboratories, Wilmington, MA) were housed and maintained as in Experiment 1. Experiment 2 was run in two replications (n = 32/replication). The free-feeding body weights at the beginning of the experiment were 330–390 g in the first replication, and 380–460 g in the second replication.

Procedure. Procedures used in this experiment were similar to those used in Experiment 1. However, prior to the 10-min pretest session, subjects in the present study were given access to sucrose to consume in their home cage over two successive nights to ensure that rats would consume sucrose in the preference apparatus from the start of the conditioning phase. In the first replication, subjects were given unlimited access to sucrose, and in the second replication, sucrose intake was inadvertently restricted to 25 ml/night. All subjects were then pretested for their baseline location preferences 2 days after their home cage exposure to sucrose.

Conditioning phase. Rats were assigned to four different groups matched for their pretest side preferences, sucrose home cage intakes, and body weights. These four groups were designated on the basis of which solution would be injected prior to each conditioning session: (a) group saline, (b) group 0.1 mg/kg naltrexone, (c) group 1.0 mg/kg naltrexone, and (d) group 5.0 mg/kg naltrexone. In addition, the sucrose-associated and water-associated locations and stimuli were fully counterbalanced within each group. Conditioning was conducted over the next 20 sessions, as in Experiment 1, except that in the present study subjects were injected with their respective treatment (1 ml/kg, SC) 15 min prior to each of the CS+ and CS− conditioning sessions.

Test phase. Testing occurred over a 2-day sequence following the final conditioning session. During these 10-min test sessions, each group was injected with saline 15 min prior to one test, and with the dosage of naltrexone that they had been conditioned with 15 min prior to the other test. Group saline was injected with 5.0 mg/kg naltrexone during their naltrexone test session. The order of saline or drug test sessions was counterbalanced across subjects in each group.

Results

Because the preference results did not vary across replications, all of the data have been collapsed across that factor.

The intake data from the conditioning sessions for each group are displayed in Fig. 2. This figure illustrates that all groups consumed more sucrose than water, but that naltrexone substantially decreased the amount of sucrose consumed. Water intakes were very low throughout the conditioning phase.

The sucrose intake data were evaluated with a group × session split-plot ANOVA. This analysis revealed significant group, $F(3, 60) = 21.52$, $p < 0.0001$, and session, $F(9, 450) = 11.98$, $p < 0.0001$, main effects, but no interaction. Rodgerian post hoc tests (33) were conducted to examine the group main effect. These tests indicated that the highest dose of naltrexone (5.0 mg/kg) decreased sucrose intake to a greater degree than the two lower doses, and that all naltrexone doses decreased sucrose intake relative to the saline group.

Figure 3 illustrates CPPs following saline and naltrexone tests for each group. All groups preferred the sucrose-associated side to the water-associated side when tested with saline

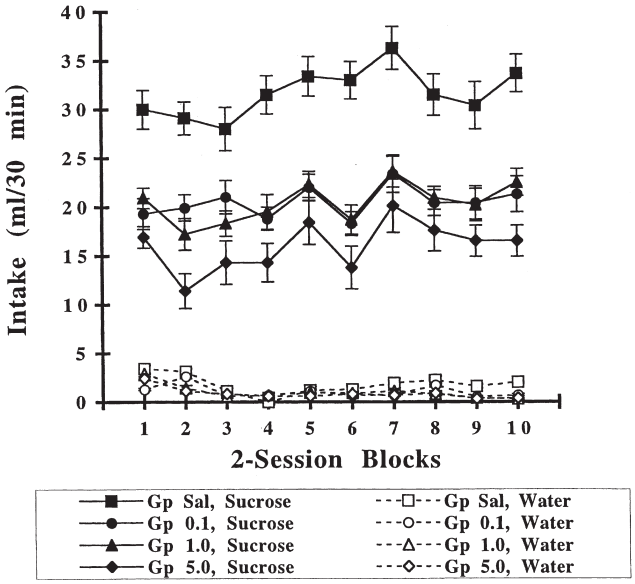

FIG. 2. Mean sucrose and water intakes (±SEM) during the 10 two-session blocks of conditioning in Experiment 2. Prior to each session, different groups of rats were injected either with saline (Gp Sal), 0.1 mg/kg naltrexone (Gp 0.1), 1.0 mg/kg naltrexone (Gp 1.0), or 5.0 mg/kg naltrexone (Gp 5.0). Naltrexone dose dependently reduced sucrose intake.

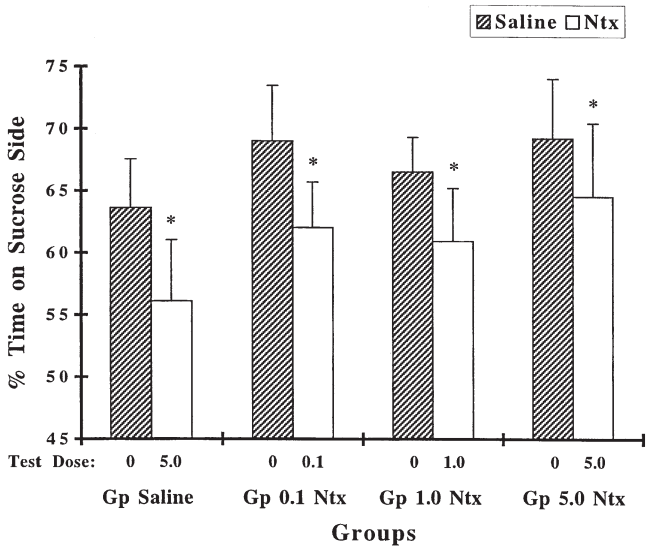

FIG. 3. Mean % time spent by each group in the sucrose-associated compartment (+SEM) during the test sessions of Experiment 2. Rats were tested after being injected with saline or the dose of naltrexone that they had received during the conditioning phase (the saline control group was tested with 5.0 mg/kg naltrexone in this test). Asterisks denote a significant main effect comparing saline and naltrexone test sessions.

(64–69%), and reduced this preference when tested with naltrexone (56–64%). A group × injection (saline or naltrexone) split-plot ANOVA performed on this data supported this claim by revealing a significant main effect of injection, $F(1, 60) = 6.97$, $p < 0.01$. Neither the group main effect nor the group × test injection interaction were significant.

An additional post hoc analysis was performed to determine if the preference scores exceeded a no-preference baseline of 50%. Separate *t*-tests comparing the preference scores in each group against 50% indicated that significant preferences were obtained in the saline tests in group saline, $t(15) = 3.48$, $p < 0.003$, group 0.1, $t(15) = 4.25$, $p < 0.0007$, group 1.0, $t(15) = 5.91$, $p < 0.0001$, and group 5.0, $t(15) = 4.00$, $p < 0.001$. During the naltrexone tests, significant preferences were displayed by group 0.1, $t(15), = 3.27$, $p < 0.005$, group 1.0, $t(15) = 2.57$, $p < 0.02$, and group 5.0, $t(15) = 2.44$, $p < 0.03$, but not by group saline.

Discussion

The present data again demonstrate that naltrexone administered during testing reduced the expression of a previously learned sucrose-reinforced CPP. The control group in the present experiment was trained very similarly to the subjects in Experiment 1, and their pattern of data was also similar. In addition, each of the three groups trained with naltrexone also expressed lower CPPs when tested with naltrexone compared to saline. This result is important in demonstrating that the deleterious effect of naltrexone on the expression of the CPP was probably not due to state-dependent learning or drug novelty effects. If these effects accounted for why naltrexone reduced the CPP in group saline, then we would not have expected naltrexone to have reduced the CPPs in the remaining groups. If anything, the opposite outcome would be predicted in these groups.

It may be worth noting that although naltrexone reduced the preference in all groups, it eliminated it only in group saline. This result is consistent with the possibility that some process in addition to naltrexone's effect on expression of the CPP may have contributed to the results. For instance, if subjects in each of the naltrexone groups became tolerant to naltrexone's effects, then incomplete reductions in preference would be expected in these groups. However, this interpretation is complicated for two reasons. First, the sucrose intake data does not indicate that tolerance occurred. Second, the lack of a significant group × test injection interaction in the preference data does not permit us to assert that naltrexone's effect differed among the groups.

It was somewhat surprising that no dose-dependent influence on the expression of the CPP was observed in the present experiment—the lowest dose (0.1 mg/kg) and the highest dose (5.0 mg/kg) of naltrexone equally impaired the expression of the CPP. Similarly, each dose of naltrexone substantially reduced sucrose intake, with only a small difference in magnitude between the high and lower doses. This is highly consistent with the increased potency of naltrexone to inhibit intake of palatable solutions in both real-feeding and sham-feeding situations (eg., 4,5,10,16,17,19,20). Perhaps if there was more of a difference in naltrexone's effects on sucrose intake in the different naltrexone conditions, there would also have been a difference in naltrexone's effects on the expression of the CPP. Nevertheless, it is noteworthy that all doses of the drug reduced the expression of a previously learned CPP. This finding suggests a role for the opioid system in modulating the expression of the sucrose-reinforced CPP.

Another important aspect of the present results is that naltrexone administered during the training phase had no measurable effect on the acquisition of sucrose-reinforced CPPs. There were no differences in the strength of the CPP among any of the groups, despite the fact that sucrose intake during training differed between the saline and drug groups. It is indeed impressive that the 5.0 mg/kg naltrexone group consumed only about half as much sucrose as the control group throughout training, yet still demonstrated a CPP that was equal to the control group.

GENERAL DISCUSSION

The present experiments were motivated by an interest in the potential involvement of the opioid system in sucrose-reinforced preferences. Towards this end, the experiments were designed to examine separately the effects of an opioid antagonist, naltrexone, upon both the acquisition and the expression of sucrose-reinforced CPPs. Moreover, in examining the effect of naltrexone upon the sucrose-conditioned place preferences, an experimental design was chosen to control for potential place–drug conditioning that might make it difficult to otherwise detect modulatory effects of the drug upon the CPP.

The results of the present experiments suggest that the opioid system is involved in sucrose-reinforced CPPs. However, naltrexone was observed to influence only the expression, and not the acquisition, of sucrose-reinforced CPPs. As mentioned above, only one experiment has examined the role of the opioid system in food-reinforced CPPs, and this study reported, in contrast to the present results, that naloxone interfered with the learning of the CPP (1). Expression was not explored in that study. The results from the Ågmo et al. (1) study are difficult to interpret, though, given the possibility that aversive effects of the drug, independent of their modulatory effects upon CPPs, could have selectively associated with

the CS+ compartment or with sucrose. These problems were better controlled in the present study by presenting naltrexone during acquisition (Experiment 2) both on CS+ and CS− days. A second difference between the two studies was that sucrose was given to the rats prior to being placed in the CPP apparatus on CS+ trials in the previous study (1), whereas, the present studies allowed sucrose to be consumed in the CPP apparatus on CS+ trials. Yet another difference between the two studies was that Ågmo et al. (1) did not use food-restricted rats in their experiment.

Regardless of how one interprets the inconsistency between the results of Ågmo et al. (1) and those of the present studies, consideration as to how naltrexone could selectively effect expression and not acquisition of a CPP may help identify the nature of the opioid influence upon CPPs. First, suppose that opioid antagonists reduce the hedonic response to palatable foods. This claim receives experimental support from the finding that ingestive taste reactivity responses to sucrose infused intraorally are reduced by naltrexone (28). Reduced sucrose intake in the groups given naltrexone during the training phase of Experiment 2 is also consistent with this suggestion. If we were to additionally assume that a positive hedonic response to sucrose is required for expression of the CPP, then the expression of the CPP should be reduced by naltrexone during a choice test. This follows from our view that the CPP is mediated by an association between the CS+ compartment and some representation of sucrose that under normal circumstances is capable of evoking a hedonic response. The result of this evocation of a hedonic response would be to elicit approach to the compartment evoking that hedonic response. In this way, the CPP can be explained, as could its reduced expression by naltrexone.

Supporting this interpretation, Perks and Clifton (29) demonstrated that expression of a food-reinforced CPP depended upon the current value of the food used to reinforce the CPP. In their experiment, a CPP was first established by selective pairings of the CS+ compartment with a 10% sucrose solution. Water was paired with the CS− compartment. Some of the rats subsequently were taught an aversion to sucrose, by pairing sucrose with LiCl in the rats' home cages. In a subsequent CPP test in which neither sucrose nor water were present, rats who had received aversion conditioning with sucrose displayed a reduced CPP. This result strongly supports the claim that during the training phase, the CS+ compartment developed an association with a representation of sucrose, and that the expression of the CPP depended upon this sucrose representation's continued ability to evoke a positive hedonic response. Other research has demonstrated that the hedonic response to a taste is, in fact, reduced by aversion conditioning [e.g., (6)].

What remains to be explained in the present experiments is why naltrexone should have influenced the expression but not the acquisition of a CPP. It seems possible that after training with naltrexone, the hedonic value of sucrose could have been restored to its normal level in the saline test compared to the naltrexone test. Consequently, the CPP should also have been restored in the saline test. But for this restored hedonic response to the sucrose representation to result in equally strong CPPs among the groups of Experiment 2, it would have to additionally be assumed that the strength of the association between the CS+ compartment and the sucrose established during training was equal in these different groups.

The claim that equally strong place–sucrose associations could have formed even if the hedonic response to sucrose differed among the groups during training is reminiscent of the old debate in the learning literature as to the necessity of reinforcement for learning to occur. Demonstrations of "latent learning" in that literature [e.g., (21)] are consistent with the dissociation claimed here. Moreover, modern treatments of conditioning theory often maintain the view that associations between neutral stimuli are readily formed (e.g., [24,31,32)], and that conditioned responses based on such associations can be present or absent, depending upon the incentive value currently assigned to the associated stimuli [e.g., 11–13)]. Thus, contemporary learning theory informs us that we might very well expect a dissociation between expression and acquisition effects of drugs upon CPPs.

It is worth noting that in Experiment 2 rats were exposed to sucrose in their home cages prior to receiving sucrose in the CPP chambers while in a drugged state. It is possible that this prior exposure to sucrose in a nondrugged state contributed to a stronger CPP in these animals when tested with saline compared to naltrexone. Other investigators [e.g., (12)] have demonstrated that experience with the reinforcer in the test drive state is necessary for an instrumentally conditioned response to be sensitive to a shift in drive from conditioning to an extinction test—an effect known in the literature as "incentive learning." It remains to be determined whether (in Experiment 2) the preconditioning phase exposure to sucrose would be necessary for obtaining the larger CPP seen in the saline test. However, it is important to realize that while this incentive learning effect has been documented for instrumentally conditioned responses, Pavlovian conditioned responses are not currently thought to require incentive learning (12).

There are at least two additional interpretations of the present finding of a dissociation between naltrexone's effects on acquisition and expression of a sucrose-reinforced CPP. First, it is possible that although naltrexone might have reduced the hedonic value of sucrose in Experiment 2 during the conditioning phase, the reduced value of sucrose was high enough to establish a strong CPP. This possibility is made less attractive by the finding that naltrexone was presumably capable of reducing the hedonic value of sucrose enough to diminish the expression of the CPP in Experiments 1 and 2. At the very least, this suggests that expression processes are more sensitive than acquisition processes to naltrexone's effects. Second, we conducted a single probe test after rats had been given 10 blocks of place preference conditioning. It remains possible that differences in the rates of acquisition might have emerged between the groups of Experiment 2.

In contrast to the present findings that naltrexone attenuated the expression of a sucrose-reinforced CPP, parallel studies demonstrated that naltrexone had little or no effect on the expression or acquisition of sucrose-reinforced flavor preferences (36,44). In these studies, rats were trained with CS+ flavors paired with either the sweet taste of sucrose (using a sham-feeding procedure) or with the postingestive actions of sucrose (using an intragastic infusion procedure). Although contrary results have been reported using other flavor conditioning procedures (23,30), taken together, our data suggest that opioid systems are involved in the expression of food-conditioned place preferences but not conditioned flavor preferences. The involvement of other neurochemical systems in these food-conditioned preferences is currently under investigation.

One remaining issue concerns the relationship between CPPs conditioned by food reinforcement and those conditioned by opiate drugs and opioid peptides. Similarities between these types of CPPs are suggested by the observations

that sucrose-induced hyperphagia alters hypothalamic opioid peptide levels (42), and that CPPs can be conditioned by morphine infusions into the lateral hypothalamus (41). Moreover, the present results are consistent with data documenting that opioid receptor antagonists interfere with place preferences conditioned by opiates and opioids [e.g., (2,3,38)]. Thus, given that opiates and opioid peptides appear to reinforce CPPs primarily through the μ, or δ, but not the κ, opioid receptor subtypes [see reviews: (15,37)], it would be of additional interest to determine which of these opioid receptor subtypes participate in the mediation of sucrose-reinforced CPPs.

In summary, the present studies demonstrate that the opioid system is involved in sucrose-reinforced CPPs. Specifically, it seems more important in the expression rather than the acquisition of this CPP. This dissociation in the effects of naltrexone upon sucrose-reinforced CPPs is consistent with the view that endogenous opioid systems are related to the hedonic value of food (7). Thus, opioid-induced modulations in the value of sucrose may be more important for performance than it is for learning processes in the CPP procedure. But regardless of the theoretical interpretation of the dissociation between acquisition and expression effects reported here, the present studies can also be viewed to be important for methodological reasons. The present study, by administering the drug on both CS+ and CS− trials during training of the CPP, seems better suited to evaluate the modulatory effects of drugs upon CPPs.

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