SUPPRESSION OF CONSUMMATORY BEHAVIOR ELICITED BY A LITHIUM-CONDITIONED FLAVOR

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by

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A Thesis submitted in partial fulfillment
of the requirements for the degree of
Doctor of Philosophy

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Memorial University of Newfoundland
January 1979

St. John's Newfoundland

Abstract

A rat develops an aversion to the flavor of a solution which is paired with illness. Although it is well established that the rat will avoid consuming this flavor, the actual conditioned response (CR) that it elicits has not been systematically investigated. The following series of experiments present a technique for measuring this CR. The rats were first trained to discriminate between an aversive flavored solution (CS+) which was paired with lithium and a safe flavored solution (CSc) which was presented alone. The CR was then measured by the ability of the immediate aftereffect of the CS+ flavor to suppress consumption of a differently flavored solution (the test solution). While the rats consumed the test solution, they were intraorally infused with 2 ml of either the CS+ or CSc flavored solution; the time to resume drinking and the subsequent rate of licking of the test solution were recorded.

All experiments demonstrated that the rats infused with the CS+ flavored solution were more hesitant to resume drinking the test solution than were the rats infused with the CSc flavored solution. This CR was evident whether the test solution was unflavored water, a novel flavored solution or a conditioned aversive flavored solution; however, the duration of the CR varied by the nature of the test solution, ranging between 45 and 235 seconds. Finally, the strength of the suppressive CR was influenced by variations of the sickness intensity and the flavor intensity during conditioning.

It is unlikely that the CR measured in these experiments is the sole motivator of a flavor aversion, because extinction of the suppressive CR did not even weaken the rats subsequent avoidance of the CS+ flavored solution. The suppressive CR elicited by a lithium-conditioned flavor parallels the suppressive CR elicited by a shock-conditioned external cue.

ACKNOWLEDGEMENTS

I would like to express my gratitude to Dr. Sam Revusky for his critical evaluation of a number of earlier drafts of this thesis. His advise and guidance have been extremely valuable throughout the course of this work. In addition, I am grateful to Dr. John Evans, Dr. Bow Tong Lett and Dr. Bill McKim for their helpful comments on earlier versions of this manuscript.

I also thank Shannon Coombes for his excellent technical assistance in the experiments reported in Appendix B, and Spence Butt for his assistance in the construction of necessary equipment. Finally, for moral support when I most needed it, I thank various friends at Memorial, but especially Svend Stouge who was always ready to listen.

This work was financially supported by the Psychology Department at Memorial and by an NRC grant awarded to Dr. Revusky. Experiments 1 and 1^a were presented at the Canadian Psychological Association Meetings, Toronto, 1976.

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CHAPTER I: INTRODUCTION

When a rat is injected with lithium chloride after drinking a flavored solution, it will drink less of that solution in the future. It is usually stated that the establishment of this learned aversion follows the principles of Pavlovian conditioning. Pavlov's (1927) stimulus substitution theory states that after a number of presentations of a neutral conditional stimulus (CS) in temporal contiguity with a biologically meaningful unconditional stimulus (US), the former gains the capacity to elicit conditioned responses (CRs) much like the unconditioned responses (URs) reflexively elicited by the US. Essentially, the CS becomes substituted for the US. In flavor aversion learning, a flavor (CS) is paired with an injection of lithium (US) which produces sickness (UR). If Pavlov's paradigm applies to flavor aversion learning, the presentation of the CS alone ought to elicit some components of the unconditioned sickness response, which may then be termed the conditioned sickness response (CR).

However, flavor aversion learning is generally not measured in terms of a conditioned sickness CR, but as an avoidance of CS flavored water. In this test of a flavor-lithium association, the rat approaches a bottle containing the CS solution and licks from the spout in order to identify the flavor of that solution. It engages in a number of such approaches during a test period, typically of several minutes. After each CS exposure, the rat may display the CR of agitated withdrawal from the spout, jerking its head backwards, grooming and rubbing its chin on the floor (Garcia, Clarke and Hankins, 1973).

The role of a sickness UR in flavor aversion learning.

Although an immense number of experiments have demonstrated learned avoidance of flavored substances (See Riley and Baril, 1977), few experiments have investigated the nature of the actual CR elicited by the flavor CS+. Not all such learned flavor aversions involve conditioned sickness, since some USes which produce flavor aversions do not produce clearcut sickness URs. If there is no sickness UR, clearly there is no basis for a sickness CR. Early studies (Garcia, Kimmeldorf and Koelling, 1955) showed that

doses of radiation well below the clinical threshold for radiation sickness were capable of producing flavor aversions. Furthermore, moderate doses of drugs which humans use for recreation, like the barbiturates, minor tranquilizers, and amphetamine, are also capable of producing flavor aversions (Gamzu, 1977). The strongest basis for doubting that a drug which is capable of producing a flavor aversion must elicit a sickness UR is the finding that amphetamine will not only serve as an aversive reinforcer to establish a flavor aversion when injected after consumption of a flavored solution (e.g., Berger, 1972), but will also serve as an appetitive reinforcer to establish increased responding when injected intravenously (IV) after a specified number of bar presses in a Skinner Box (e.g., Pickens and Harris, 1968). In fact, this dichotomous effect has been demonstrated in rats which were injected intraperitoneally (IP) with amphetamine immediately after they consumed a flavored solution in a distinctive location; in subsequent tests, the animals both avoided the flavored solution and approached the distinctive location (Reicher and Holman, 1977). Apparently, "the amphetamine state" is aversive when paired with drinking and rewarding when paired with motor responses (See also Wise, Yokel and De Witt, 1976). For

this reason, the internal state elicited by an injection of amphetamine cannot be characterized simply as sickness even though it produces a flavor aversion.

Although it is not tenable to attribute all instances of conditioned flavor aversions to conditioned sickness, it is quite certain that many USes which produce flavor aversions also produce sickness. Lithium chloride, a common US in flavor aversion learning experiments, directly affects the gastrointestinal tract producing nausea, salivation, vomiting, and diarrhea in man and animals (Schou, 1968; Boland, Mellor and Revusky, 1978). Animals which are sacrificed after a lethal dose of lithium have been shown to have a "marked hyperemia and hypermotility of the stomach and small intestine with the entire gastrointestinal tract distended with fluid" (Davenport, 1950). Since lithium elicits a UR of gastrointestinal distress, stimulus substitution theory would suggest that a flavor paired with lithium will elicit a conditioned sickness response.

Evidence for a classical conditioned sickness response.

A conditioned sickness CR elicited by an external CS was initially observed and directly

measured by Pavlov (1927) as is indicated in the passage below:

> "A dog was given a small dose of apomorphine subcutaneously and after one or two minutes a note of a definite pitch was sounded during a considerable time. While the note was still sounding the drug began to take effect upon the dog; the animal grew restless, began to moisten its lips with its tongue, secreted saliva and showed some disposition to vomit. After the experimenter had reinforced the tone with apomorphine several times it was found that the sound of the note alone sufficed to produce all the active symptoms of the drug, only in a lesser degree."

More recently, Pavlov's paradigm has been used to study a "conditioned withdrawal" CR in morphine-dependent animals. Morphine withdrawal is characterised by symptoms of sickness such as excessive salivation, vomiting and body temperature changes; these symptoms have been conditioned to telereceptive (external) cues (Irwin and Seevers, 1956; Wikler, 1965; Goldberg and Schuster, 1970). Morphine dependent animals are withdrawn from morphine or are administered the morphine antagonist nalorphine to induce withdrawal, i.e. the US. The withdrawal syndrome either occurs in a specific environment or in the presence of a distinctive environmental cue, i.e. the CS. After a number of such pairings, the CS alone elicits the symptoms of withdrawal, even when presented months later when the animals are no longer

morphine dependent (Irwin and Seevers, 1956; Goldberg and Schuster, 1970).

The studies reported above show that conditioned sickness responses have been established to telereceptive CSes, even though the physiological sickness reactions are slow in onset and may not occur for minutes after the CS presentation. Since taste and related stimuli are more easily associated with interoceptive consequences than are telereceptive stimuli (e.g., Garcia and Koelling, 1966), it is reasonable to assume that such CSes will effectively support a conditioned sickness response.

Evidence of a conditioned sickness response in flavor aversion learning.

Sickness CRs have been conditioned to flavors. Zahorik (1972) demonstrated that physiological changes characteristic of thiamine deficiency are capable of becoming conditioned to a flavor CS. While in a thiamine deficient state, which causes physiological changes which include decreased heart rate, rats consumed a distinctively flavored solution (CS). They were later presented the CS flavor, but in a non-deficient state, and showed a decreased heart rate CR. The flavor CS had gained the capacity to

elicit at least one component of the unconditioned sickness response.

Since rats are incapable of vomiting, they do not display obvious behavioral evidence of conditioned sickness when presented a flavor previously paired with lithium; however, a clearcut sickness CR, which resembles a lithium-induced sickness UR, has been shown in other species. When confronted with their conditioned aversive prey, coyotes, cougars and ferrets retch (Gustavson, Kelley, Sweeney and Garcia, 1976; Rusiniak, Gustavson, Hankins and Garcia, 1976) and Buteo hawks vomit, smack their beaks and engage in head flipping which is characteristic of the sickness UR (Brett, Hankins and Garcia, 1976). In addition to their obvious sickness CRs, a number of species, including the rat, display disgust reactions to conditioned aversive substances, such as emptying food cups, grooming themselves and rubbing their noses along the bottom of the cages, which may be similar to behaviors associated with illness (Gustavson, 1977).

Aside from observational evidence, there is recent pharmacological evidence which suggests that a flavor previously paired with illness elicits a

a sickness CR. Coil, Hankins, Jenden and Garcia (1978) have reported that an aversion to a flavor previously paired with lithium may be disrupted by pretreatment with an antiemetic drug. Rats that were injected with a low dose of an antiemetic agent thirty minutes before a drinking test consumed more of their CS flavored solution than rats that were injected with saline. Presumably, the drug suppressed the emetic mechanisms that are normally activated by exposure to the CS flavor.

The above studies strongly suggest that a flavor paired with lithium-induced sickness gains the capacity to elicit a conditioned sickness response. However, since a flavor-illness association is generally demonstrated as an avoidance response in which a rat controls its own exposure to the CS flavor by approaching and withdrawing from a bottle containing the solution, the actual sickness CR elicited by the flavor CS has not been systematically investigated.

Conditioned Suppression of drinking: A measure of conditioned sickness.

A technique by which to measure conditioned sickness has been devised by Green, McGowan and Garcia

(as reported in Garcia, McGowan and Green, 1972). They reasoned that since rats which experience a sickness UR selectively suppress their consumption of novel flavored solutions (Green, McGowan, Garcia and Ervin, 1968; Domjan, 1977), rats which experience a sickness CR should also show such suppression. They found that rats with a history of four apomorphine injections respond to an injection of isotonic saline (CS) by suppressing their intake of a novel saccharin solution for the first four minutes of a 10 minute drinking period. As already indicated, gustatory cues are more associable with illness than are nongustatory cues (e.g., Garcia and Koelling, 1966; Domjan and Wilson, 1972). For this reason, an illness-paired flavor CS would be expected to elicit a stronger sickness CR than would an injection procedure. The following series of experiments employ a new technique for measuring the CR elicited by a lithium-paired flavor CS which resembles that of traditional conditioned suppression of licking (Leaf and Muller, 1965; Leaf and Leaf, 1966).

Table 1 compares the traditional conditioned suppression paradigm which measures a conditioned

TABLE 1.

CONDITIONED SUPPRESSION OF LICKING

	Conditioned Emotional Response	Conditioned Sickness Response
Phase 1	Train to lick from tube	Train to lick from tube
Phase 2	Tone CS → Shock US	Flavor CS → Lithium US
Phase 3	Suppression Test: Present tone CS while rats are licking the Test Solution	Suppression Test: Present flavor CS while rats are licking the Test Solution*
Results	Suppression of licking following (or during) tone	Suppression of licking following flavor

* In Experiment 1, the flavor CS was presented prior to presentation of the Test Solution instead of during its presentation.

emotional response (CER) with the paradigm of the following series of experiments which measures a conditioned sickness response. In traditional conditioned suppression of licking, rats suppress their drinking of a test solution while they are exposed to a stimulus, for example, a tone, which has previously been paired with shock. The tone CS+ elicits a CER which is measured by suppression of ongoing behavior. More recently, the strength of the CER has been measured by the duration of the suppressive effect following the termination of the CS+ (Tenen, 1967). While rats licked from a tube of sucrose solution, they were exposed for three seconds to a tone (CS+) which had previously been paired with shock; immediately after the CS+ was terminated, the time required to accumulate 3 seconds of drinking measured the strength of the CER. The following experiments employed a procedure similar to that of Tenen (1967), but a flavor was the CS and lithium was the US. The conditioned sickness response was measured by the immediate aftereffect of an exposure to a flavor CS+, which predicted illness, on the consumption of a different test solution.

CHAPTER II: BASIC CONDITIONED SUPPRESSION EFFECT

Experiment 1

Experiment 1 tested for conditioned suppression of novel fluid consumption following a brief exposure to a lithium-paired flavor CS+. All rats were subjected to a discrimination conditioning procedure in which one flavor was paired with lithium (CS+) and the other flavor was paired with saline (CSc). The CSc group measured the response elicited by an equally familiar, but safe, flavor. The flavor which served as CS+ was counterbalanced in order to ensure that the suppressive effect was general across CS flavors; for half the rats the CS+ was vinegar and the CSc was coffee, and for the remaining rats, the roles of the flavors were reversed. On the test day, the rats were exposed to the CS+ flavor, the CSc flavor,or unflavored water immediately before a 5-minute presentation of novel sucrose solution.

Method

Subjects. Sixty male Sprague-Dawley rats, ranging between 129 - 183 gms, individually housed in stainless steel cages, were maintained on ad-lib

rat chow throughout the experiment. All experimental manipulations except weighings, injections, and infusions were conducted in their home cage.

Flavored solutions. Three novel flavored solutions were used: 1.25% (w/v) Sanka decaffeinated coffee, 3% (v/v) Heinz cider vinegar and 15% (w/v) sucrose, all mixed with tap water. The coffee and vinegar served as the discriminative stimuli, counterbalanced for their role as CS+; that is, for half of the subjects coffee was the CS+, for the other half vinegar was CS+. The solution not used as CS+ had the role of CSc. Sucrose served as the test solution (TS) which was consumed by all rats on the Test Day.

Pre-training.(Days 1-4). The rats were initially adapted to the passive infusion manipulation described below, which would later serve as the CS exposure procedure. On each of four days, the experimenter removed each rat from its home cage, placed the tip of a plastic syringe in its mouth, and infused 5 cc of water over its tongue in the course of 15 seconds. Immediately after the passive infusion manipulation, each rat

was replaced in its home cage and presented a bottle of water for a 10 minute drinking period. Six hours later they were given 10 additional minutes of water.

Discrimination Conditioning. (Days 5 & 6). The subjects were given discrimination conditioning trials on the following two days. On one day they consumed the CS+ flavor which was followed by a 16 ml/kg injection of .15M Lithium Chloride (LiCl) and on the other day they consumed the CSc flavor which was followed by an equivolume injection of isotonic saline: Half of the rats received the CS+ trial on Day 5 and the other half received it on Day 6.

On each trial, a rat was administered a 5 cc passive infusion of water over a 15 second period and was then presented a bottle containing the appropriately flavored solution. After a 10 minute drinking period, the bottle was removed and the rat was injected intraperitoneally (IP) with either lithium or saline. On the following day, the rat was given the alternate treatment. Six hours after each conditioning trial, all rats were given 10 minutes access to water in a bottle.

Baseline Training and Testing. (Days 7-10). On Days 7 and 8, all rats were adapted to a drinking schedule in which a 5 ml passive infusion of water

immediately preceded a 5 minute presentation of water in a weighed bottle. Six hours later they were presented water again for 10 minutes.

The testing procedure on Day 9 was identical to that of baseline training except that the solutions differed. Each rat was infused for 15 seconds with 5 ml of CS+ (n=20), CSc (n=20) or water (n=20) immediately before a 5 minute presentation of novel 15% (w/v) sucrose test solution (TS) and the amount of sucrose consumed was measured. Six hours later the rats were allowed to drink water for 10 minutes from a bottle.

To determine whether the test experience modified their subsequent preference for sucrose, on the following day, Day 10, the rats were given a 10 minute two-bottle choice test between sucrose and water in which sucrose was always presented on the righthand side of the cage.

Design. The infusion condition (CS+, CSc or Water) was the factor of theoretical interest; however, two additional control factors, flavor infused (coffee or vinegar) and order of CS+ training (Day 5 or 6) completed the 3 x 2 x 2 design. Individual comparisons between groups were performed by Newman-Keuls tests.

Results and Discussion

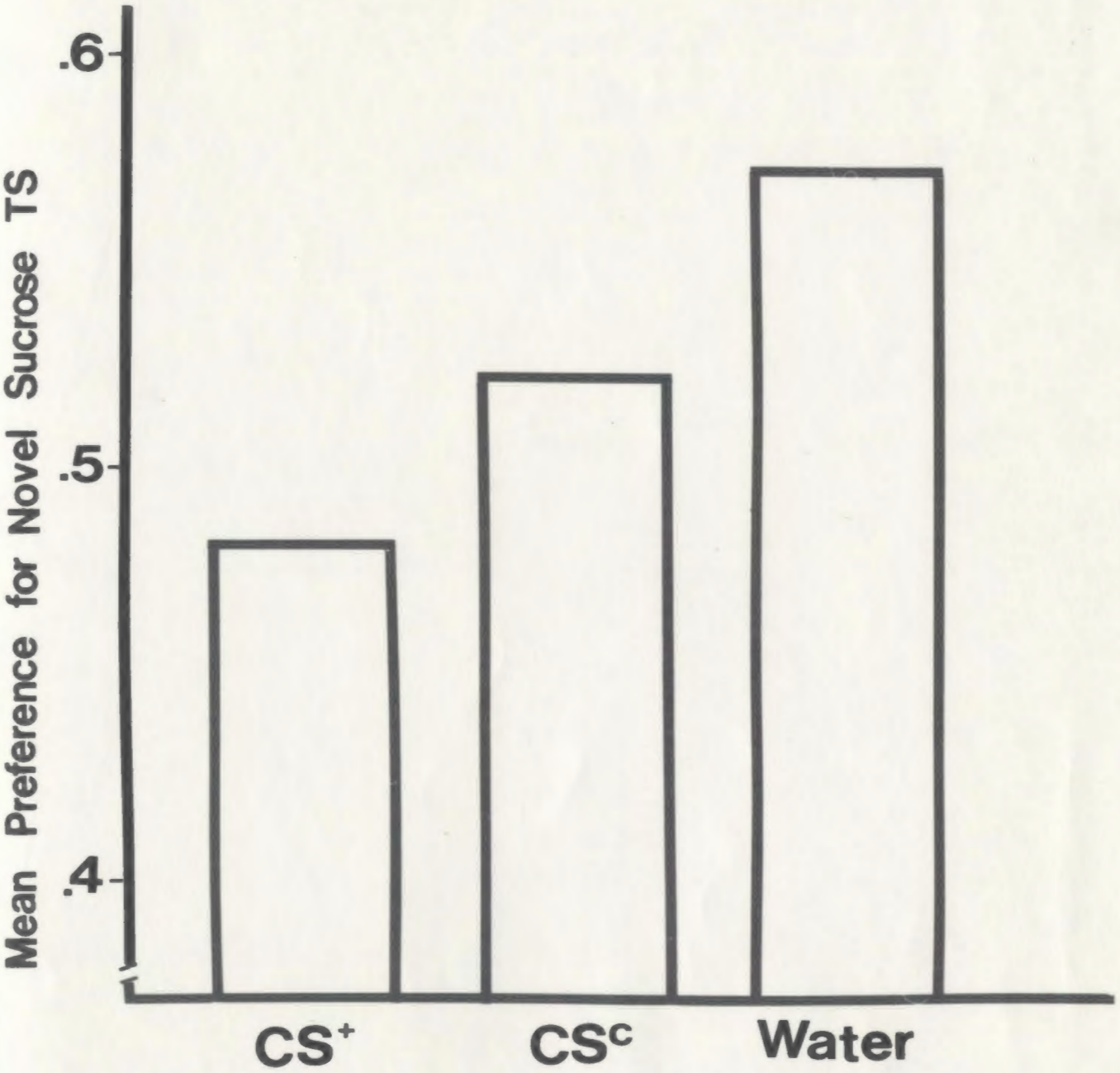
The sucrose intakes on the Test Day, Day 9, were converted to preference ratios relative to the rat's water intake on the previous day: Its sucrose intake on Day 9 was divided by the sum of its water intake on Day 8 and its sucrose intake on Day 9. A 3 x 2 x 2 ANOVA, presented in Table 2, performed on these preference ratios, revealed a significant effect of infusion condition ($F_{(2,48)} = 4.76$; $p < .025$); no other effects were significant. Since the specific flavor infused did not influence the preference for sucrose, the flavors were pooled to represent infusion condition in Figure 1. Figure 1 shows that rats which were infused with the CS+ flavor (M= .48) had lower sucrose preferences ($p < .01$, by Newman-Keuls analysis) than those which were infused with water (M= .57); but, the rats infused with CSc (M = .52) did not differ from either of the other two groups. The lack of a difference between Groups CS+ and CSc indicates that exposure to CS+ did not elicit a very marked CR; however, later experiments will show that under more sensitive conditions a lithium-paired flavor CS+ consistently elicits a suppressive CR in comparison to a CSc.

The preference for sucrose on Day 10, relative

TABLE 2. Analysis of Variance Summary Table of Sucrose preferences in Experiment 1.

Source of Variance	df	MS	F value	p
A (Infusion Condition)	2	.0451	4.764	.025
B (Flavor infused)	1	.0081	.930	
C (Order of conditioning)	1	.0057	.006	
AB	2	.0097	1.115	
AC	2	.0263	3.063	
BC	1	.0046	.526	
ABC	2	.0056	.644	
S	48	.0087		

Figure 1. Mean preference for a novel sucrose solution which was presented immediately after a passive infusion of CS+, CSc or Water in the suppression test of Experiment 1.


Mean Preference for Novel Sucrose TS
.6
.5
.4
CS+
CSc
Water

to the preference for water in the two-bottle test, was not influenced by its previous pairing on Day 9 with CS+, CSc, or water ($F_{(2,57)} = 1.33$).

Experiment 1a

Experiment 1 showed that a brief exposure to a flavor (CS+) previously paired with sickness disrupted consumption of a novel sucrose solution more than did a brief exposure to unflavored water. Unfortunately, it was not clear if CS+ disrupted drinking more than did CSc, an equally familiar flavor that was not paired with sickness, although there was an insignificant trend in this direction. In general, the magnitude of the suppression obtained in Experiment 1 was disappointingly small and a larger effect was needed if sickness CRs were to be systematically studied. In Experiment 1a, the procedure was modified in two ways in order to obtain more sensitive results: 1) the drinking test was reduced from five minutes to three minutes and 2) the test solution was changed from sucrose to unflavored water. The rationales for these changes follow.

The reduction of the drinking test period was based on the possibility, suggested by the work of Garcia, McGowan and Green (1972), that conditioned sickness effects are short-lasting. In designing Experiment 1, I supposed that this generalization would not apply if the CS+ were a flavor, since

flavors have a strong proclivity to become associated with sickness (Garcia and Koelling, 1966) and also might have longer lasting traces than other stimuli (Krane and Wagner, 1975, but see Lavin, 1976). Hence, I did not consider a five minute test period too long; but the marginal results of Experiment 1 suggested that I might be wrong.

The change in the test solution from sucrose to unflavored water was based on the possibility that the strong tasting sucrose test solution had, in some manner, masked the suppressive CR elicited by the flavor CS+. Unflavored water was expected to result in less interference.

Method

The same subjects of Experiment 1 were regrouped by infusion condition so that no animal received the same test treatment it had previously received. For instance, rats which were infused with CS+ in Experiment 1 were equally divided so that half were infused with CSc and half were infused with water on the test day of Experiment 1a. The scores were analyzed for any residual effects from the test treatment in Experiment 1.

On each of three days (Days 11, 12, and 13) beginning the day after the preference test of Experiment 1, the rats were trained to drink from two successively presented bottles for a combined total period of eight minutes. This provided two measures of amount consumed. A 5 ml passive infusion of water immediately preceded presentation of one bottle of water for 3 minutes followed by another bottle of water for an additional 5 minutes. Six hours later, the rats were given 10 minutes of water from a bottle.

The baseline training procedure also served as the test procedure except that different flavors were used. On the Test Day (Day 14), the rats were given a 5 ml passive infusion of CS+, CSc or Water over the course of 15 seconds immediately before a 3 minute presentation of unflavored water test solution in one bottle, followed by a 5 minute presentation of a novel 1.5% (w/v) Salt test solution in another bottle. The Salt test was included to measure any residual effects of the CR in the event that the suppressive effect is specific to novel test solutions. Since the Salt test was the final test, it would not effect the results of the earlier

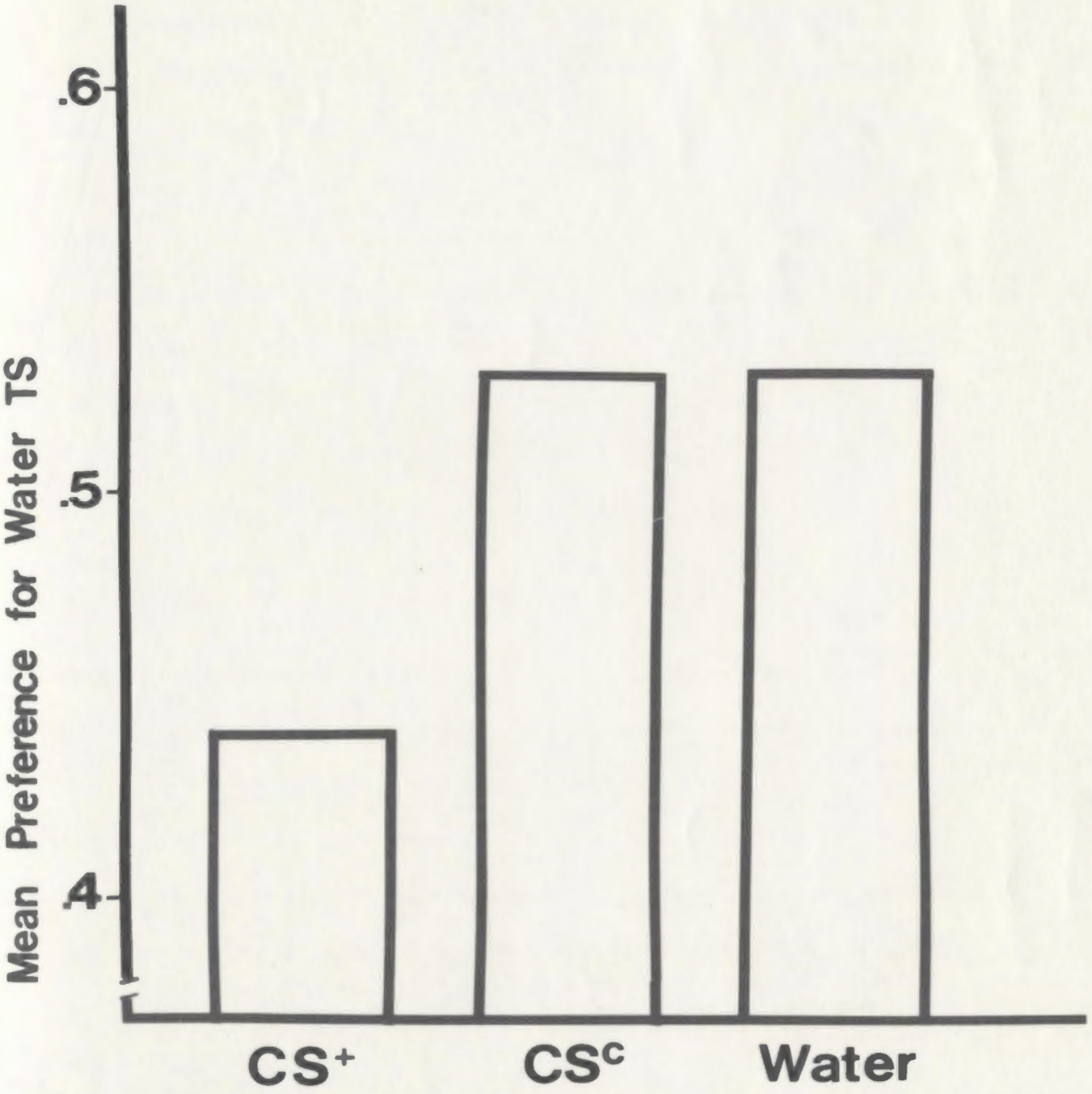
water test. The amounts consumed were recorded on all days.

Results and Discussion

Separate preference ratios were computed for the water scores and the salt scores on the Test Day (Day 14). The water preference ratios were computed relative to the first 3 minute drinking period on the preceding water day and the salt preference ratios were computed relative to the second 5 minute drinking period on the preceding water day. The factor of previous experience from Experiment 1, analyzed by individual t-tests for each infusion condition in Experiment 1^a, had no effect on the test solution preference (t's (18)$<$1.4); therefore, the scores were pooled for the remaining analyses.

As is evident in Figure 2, exposure to the CS+ flavor suppressed water consumption ($F_{(2,57)}$=12.0; $p<.01$); Newman-Keuls tests revealed that the animals which were infused with CS+ (M=.44) drank significantly less than those infused with either CSc (M=.53) or water (M=.53) at the .01 level. A one-way ANOVA performed on the subsequent five minute salt preferences, however, revealed no significant effect

Figure 2. Mean preference for unflavored water which was presented immediately after a passive infusion of CS+, CSc or Water in Experiment 1a.


Mean Preference for Water TS
.6
.5
.4
CS+
CSc
Water

of infusion condition ($F_{(2,57)}$ 1.0).

General Discussion

Exposure to a CS+ flavor which predicted illness suppressed drinking of unflavored water in a three minute drinking test. This finding is surprising in light of previous reports that the sickness UR elicited by either nitrogen-mustard (Green, McGowan, Garcia and Ervin, 1968) or lithium chloride (Domjan, 1977) selectively suppresses novel fluid consumption, but does not influence the intake of unflavored water in a 30 - 60 minute drinking test. However, Haroutunian, Riccio and Gans (1976) have recently reported that the sickness UR induced by prior rotation suppresses intake of unflavored water when measured as the latency to begin licking. Their results suggest that the unconditioned suppressive effect of sickness on water drinking may be very brief and thus may not be detectable in the typical 30 - 60 minute test. In fact, Experiment A, reported in Appendix A, showed that when a 5 or 10 minute drinking test is used, lithium-induced illness suppresses consumption of unflavored water.

Although a stronger suppressive effect was demonstrated in Experiment 1^a with unflavored water TS in a three-minute test than in Experiment 1 with novel sucrose in a five-minute test, both experiments suggested that the CS+ exposure caused greater suppression than the CSc exposure. When the results of the two experiments were pooled, the CS+ condition elicited a greater suppressive CR than did the CSc condition ($t(78) = 2.53$; $p < .01$).

Since both the nature of the test solution and the duration of the drinking period in Experiment 1^a were different than in Experiment 1, it is impossible to determine which factor was responsible for the stronger CR apparent in Experiment 1^a. Therefore, in Experiment 2, I used a novel TS (as in Experiment 1) and a new technique for measuring changes in the CR strength over time. If the five-minute drinking test of Experiment 1 was too long to detect a conditioned sickness response, then the new technique used in Experiment 2 should demonstrate the CR because it measures early effects of the CS+ exposure during a drinking test.

CHAPTER III: A NEW TECHNIQUE WHICH MEASURES CHANGES IN THE CR STRENGTH OVER TIME

Experiment 2

Experiment 1[a] suggested that the immediate aftereffect of an exposure to a conditioned aversive flavor (CS+) caused rats to drink less of another solution (TS), than did the immediate aftereffect of an exposure to a safe, but equally familiar, flavor (CSc). Although the measure of overall amount consumed within a specified time demonstrated the presence of conditioned suppression, it did not measure changes in CR strength over time. It is likely that the sickness CR is stronger immediately after CS+ presentation than at the end of the test period. Therefore, a shorter drinking interval may be a more sensitive measure of the CR than a longer interval. In order to determine the duration of the suppressive CR which follows exposure to a lithium-paired flavor CS+, in Experiment 2, the rats drinking response was monitored by means of a drinkometer apparatus in a method which closely approximated the paradigm of traditional conditioned

suppression of licking.

The basic procedures of Experiment 2 were modelled on those of traditional conditioned suppression of licking. In the traditional conditioned suppression paradigm, it is unnecessary to handle the rat during the CS+ presentation; for example, the rat is presented a tone CS+ while it licks the test solution from a tube. However, in Experiment 1, the rats were lifted from their home cages, were forcibly infused with the CS+ flavored solution and were then returned to their cages prior to the presentation of the test solution. It is conceivable that this extensive handling of the rats during the CS presentation served as a source of interference during the suppression test. Therefore, in Experiment 2, as well as in the experiments which follow, a method of CS exposure was used which did not require handling of the rats: The CS flavored solutions were presented through permanently implanted intraoral cannulae. In this new test of conditioned suppression, the rats were allowed to consume the test solution for a brief period (30 seconds) before CS+ presentation. While the rats were licking from the bottle containing the TS, they were intraorally infused with CS+ and

the time to resume licking and the rate of licking were measured.

The procedures of Experiment 2 are outlined in Table 3. The rats were first given discrimination training in which the CS+ flavor was paired with lithium on Days 4 and 6 and the CSc flavor was presented without any subsequent injection on Days 5 and 7. Then, after two days of baseline training, to adapt the rats to the test procedure, the effect of CS exposure on consumption of the novel flavored test solution was assessed. The rats were allowed to consume novel HCl solution in a bottle for 30 seconds before being intraorally infused for 15 seconds with CS+ or CSc. Immediately following the infusion, a drinkometer system was activated which provided two measures of conditioned suppression: 1) the latency to complete 10 licks and 2) the number of licks completed during each 30 second interval for 12 minutes. These measures monitored changes in the CR strength over time.

Method

Subjects. Twenty male Sprague Dawley rats ranging between 219-242 gms were treated as in Experiment 1 except as specified.

TABLE 3. The basic procedures of Experiment 2.

Phase 1		
(Discrimination Training)	CS+ → Sickness	(Days 4 and 6)
	CSc alone	(Days 5 and 7)
Phase 2		
(Suppression Test)	CS+ or CSc infusion TS → CR Measures: Time to complete 10 licks Number of licks per 30 sec.	

Flavored solutions. Two palatable novel flavored solutions were selected as discriminative stimuli: A 15% (w/v) sucrose solution and a 1.5% (w/v) NaCl solution, counterbalanced for their role as CS+. The solution not used as CS+ served as CSc. A novel flavored 1.5% (v/v) HCl solution which was orthogonal (did not generalize) with sucrose and relatively orthogonal with NaCl was the test solution. The flavors were selected on the basis of the stimulus generalization experiments reported in Appendix B.

Surgery. All rats were surgically implanted with intraoral cannulae constructed of the following materials: a 4 inch length of polyetheline 90 tubing, a 20 ga. plastic adapter cap and a 5 mm diameter plastic washer.

The surgical procedure was similar to that devised by Domjan and Wilson (1972). After being deprived of water for 24 hr., each rat was administered an initial dose of 42 mg/kg of sodium pentobarbital and supplemental doses of 15 mg/kg until it reached the required depth of anesthesia. The procedure for implantation was as follows: A 15 ga. thin wall stainless steel needle was inserted through the rat's skin in the mid-neck region,

brought subcutaneously behind its ear, along the inside of its cheek, and exited through the soft part of its cheek behind the first molar; the skin around each of the punctured sites was swabbed with alcohol. With the needle in place, a 4 inch length of P.E. 90 tubing was inserted through the barrel. The needle was then removed and the tubing was secured at the neck by a 20 ga. intramedic adapter and in the mouth by a 5 mm plastic washer. The rat was then returned to its home cage, wrapped in a paper towel to maintain its body heat. During recovery from surgery, all rats had two days of free access to water, and on the final free access day, their cannulae were flushed with water to prevent stoppage from food.

Cup Drinking Training (Days 1-3). On the third day after surgery and on each of the following three days, each animal was presented a stainless steel cup containing water for two ten-minute periods each day; the first presentation was in the morning when the rat was 18 hr. water deprived and the second was 6 hr. later. This deprivation schedule was maintained throughout the remainder of the experiment.

Discrimination Conditioning Trials. (Days 4-7). On the next four days, the rats were given discrimination conditioning trials. On Day 4, each rat was presented a cup containing 10 ml of its CS+ flavored solution for 10 min.; ten rats drank sucrose and ten rats drank NaCl. Immediately after the cup was removed, the rat was injected with 1.5 ml of 2% (w/v) LiCl in solution with distilled water. On Day 5, each rat was presented the same amount of the alternate flavored solution (CSc) as it had consumed of the CS+ flavor on Day 4 but was not injected after removal of the cup. The conditioning procedure of Days 6 and 7 was similar to that of Days 4 and 5 respectively, except that the dose of LiCl administered on Day 6 was increased to 2.5 ml. If a rat consumed less than 3 ml of the CS+ solution on Day 6, it was given a 2 ml passive infusion (as described in Experiment 1) of this solution prior to the lithium injection and on Day 7 was also given a 2 ml passive infusion of the CSc solution. Therefore, each subject consumed only as much of the CSc solution as it had consumed of the CS+ solution.

Six hours after each conditioning trial, the subjects were allowed to quench their thirst in

an additional 10 minute drinking period; the water was presented in cups in order to extinguish any possible association that may have developed between the cup and toxicosis in the initial CS+ trial. Six hours after the final conditioning trial, the cannulae were flushed and all rats received 18 hr. access to water in a bottle.

Baseline Training Trials (Days 9 - 10). On Days 9 and 10, the rats were given baseline training sessions. The food was removed from each rat's home cage 30 minutes before each session began and was replaced 30 minutes after the end of a session. At the beginning of a session, a 15 inch infusion hose was connected to the adapter of each animal's cannula and a syringe containing water was attached to the end of the hose outside of the home cage. The rat was then presented a bottle of water with a spout connected to a drinkometer relay system which recorded each lick. The spout of the bottle was constructed of glass with a 2 mm opening at the end distal to the rubber stopper, and, in order to prevent electrical short circuits in the system, was covered with plastic which protruded 1/4 inch beyond its tip.

Thirty seconds after each rat began to drink from the bottle, it was intraorally infused with 2 ml of water over the course of 15 seconds. Immediately after the infusion, the drinkometer system was activated which started two clocks: One clock timed the latency to lick the first 10 licks and the other clock timed 30 second intervals for 12 minutes after presentation of the flavor. Thus the time to complete 10 licks and the number of licks per 30 second interval were recorded. Following every trial, the spouts were thoroughly dried to prevent electrical short circuits in the system. On each of these baseline training days, the rats were given two trials: One in the morning when water deprived for 18 hr. and the other 6 hr. later.

Suppression Test (Day 11). The testing procedure was identical to that of baseline training in unspecified details. On Day 11 when 18 hr. water deprived, the rats were presented a bottle containing a novel 1.5% (v/v) normal HCl test solution; thirty seconds after they began to drink, the rats were infused, over the course of 15 seconds, with 2 ml of either the CS+ flavor

or the CSc flavor with which they were previously conditioned. Immediately after the infusion, the drinkometer system was activated which recorded the latency to lick 10 licks and the rate of licking per 30 second interval for 12 minutes.

Design and Data Analysis. The question of theoretical concern was whether CS+ would produce greater suppression than CSc, while the counterbalanced control factor of flavor infused completed the 2 x 2 design. Although there were originally five subjects per group, during the course of the experiment three animals lost their cannulae so that there were four subjects in Groups Sucrose CS+, Sucrose CSc and NaCl CSc.

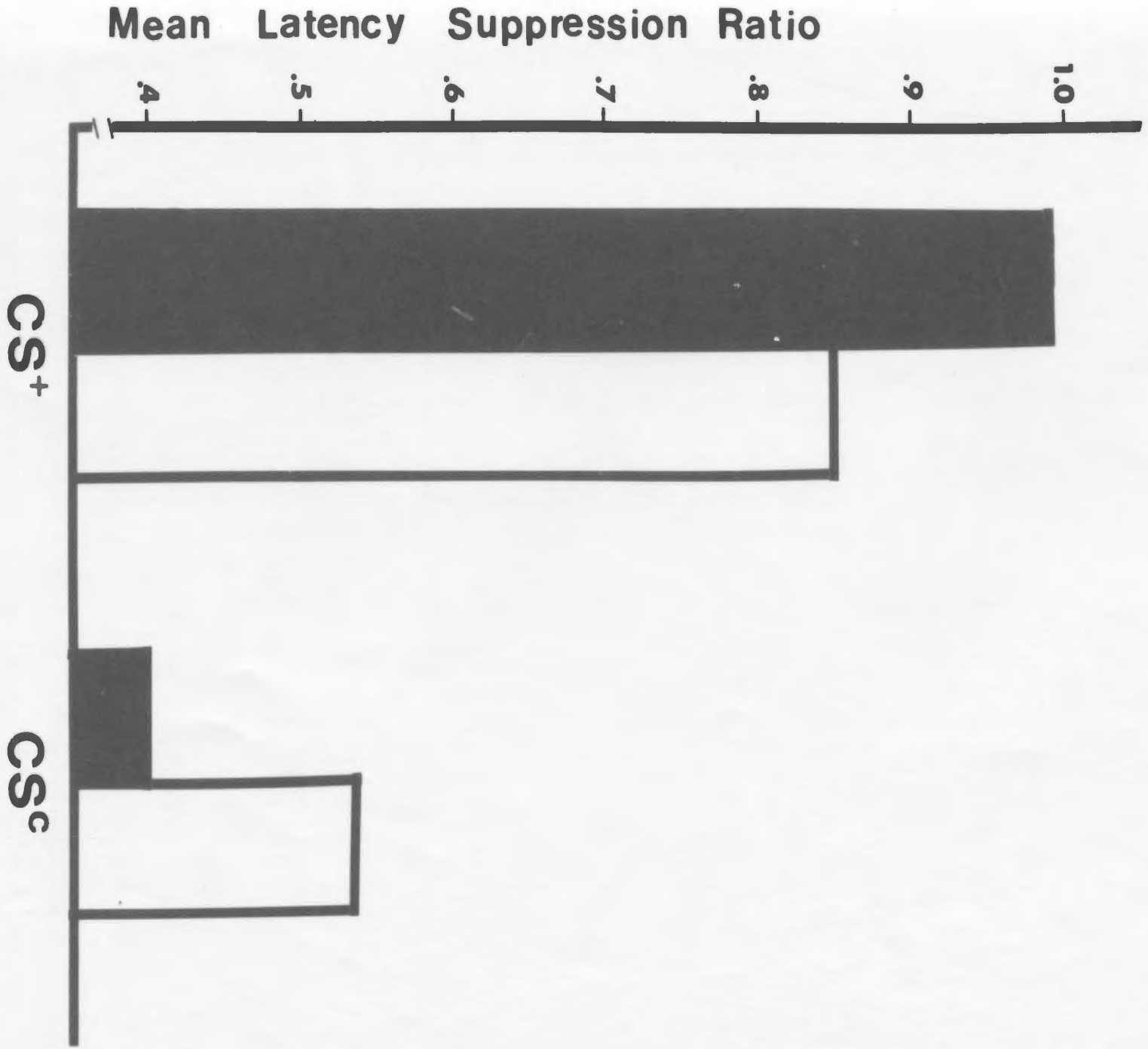
In order to control for individual differences in baseline responding, both the latency and the lick rate scores were transformed into suppression ratios (SRs) relative to the measure taken on the previous baseline day. A latency SR represented the rat's test day latency score divided by the sum of its latency score on the test day and on the preceding baseline day. A value higher than 0.5 indicated a suppressive effect. These latency SRs were analyzed in a 2 x 2 unweighted means ANOVA.

The number of licks completed in each one minute licking interval was also transformed into a suppression ratio: The number of licks on the test day was divided by the sum of the licks on the test day and the licks during the corresponding interval of the previous baseline training day. A value lower than 0.5 indicated a suppressive effect. In order to comprehensibly compare changes in drinking tendencies over time, the mean lick rate SR in each three minute block of licking was initially used as input in an ANOVA. These scores were analyzed in a 2 x 2 x 4 unweighted means repeated measures ANOVA and in a subsequent trend analysis by the method of orthogonal comparisons.

Results and Discussion

Latency Measure. Figure 3 presents the mean latency SR for each group in Experiment 2. The pair of bars to the left are the CS+ conditions and the pair of bars to the right are the CSc conditions. Within each pair, the closed bar is sucrose and the open bar is NaCl. Obviously, the CS+ groups were more hesitant to resume licking the novel HCl test solution than were the CSc

Figure 3. Mean Latency suppression ratio $\left(\frac{\text{Test Day Latency}}{\text{Test Day Latency + Baseline Latency}}\right)$ for each group in Experiment 2. The closed bars represent Sucrose and the open bars represent NaCl.


Mean Latency Suppression Ratio
.4
.5
.6
.7
.8
.9
1.0
CS+
CSc

groups ($F_{(1,13)}=66.23; p<.01$). Although the infusion condition x flavor of infusion interaction was also significant ($F_{(1,13)}=6.20; p<.05$), Newman-Keuls comparisons between conditions revealed no differences. Furthermore, the specific flavor infused, in itself, did not effect the latency to lick ($F_{(1,13)}= 0.002$).

Lick Rate Measure. The mean lick rate SR for each minute following either the CS+ or CSc infusion is presented in Figure 4. Although the analysis, shown in Table 4, revealed no overall significant effect of infusion condition ($F_{(1,13)}=.12$), it did reveal a significant infusion condition x intervals interaction of the type which would be expected if conditioned sickness effects dissipate over time ($F_{(3,39)}=2.32; p<.05$, one-tailed). The trend of licking by Groups CS+ and CSc differed across the four three-minute blocks of licking ($F_{(1,39)}=3.53; p<.05$, one-tailed). In the first three minute block, Group CS+ showed significantly greater suppression of licking than did Group CSc ($t(15)=2.23; p<.025$). Since the CR was expected apriori to be strongest during the early phase of the test period, each of the first three minutes was

Figure 4. Mean lick rate suppression ratio for CS+ and CSc conditions across the twelve minutes of licking in Experiment 2.

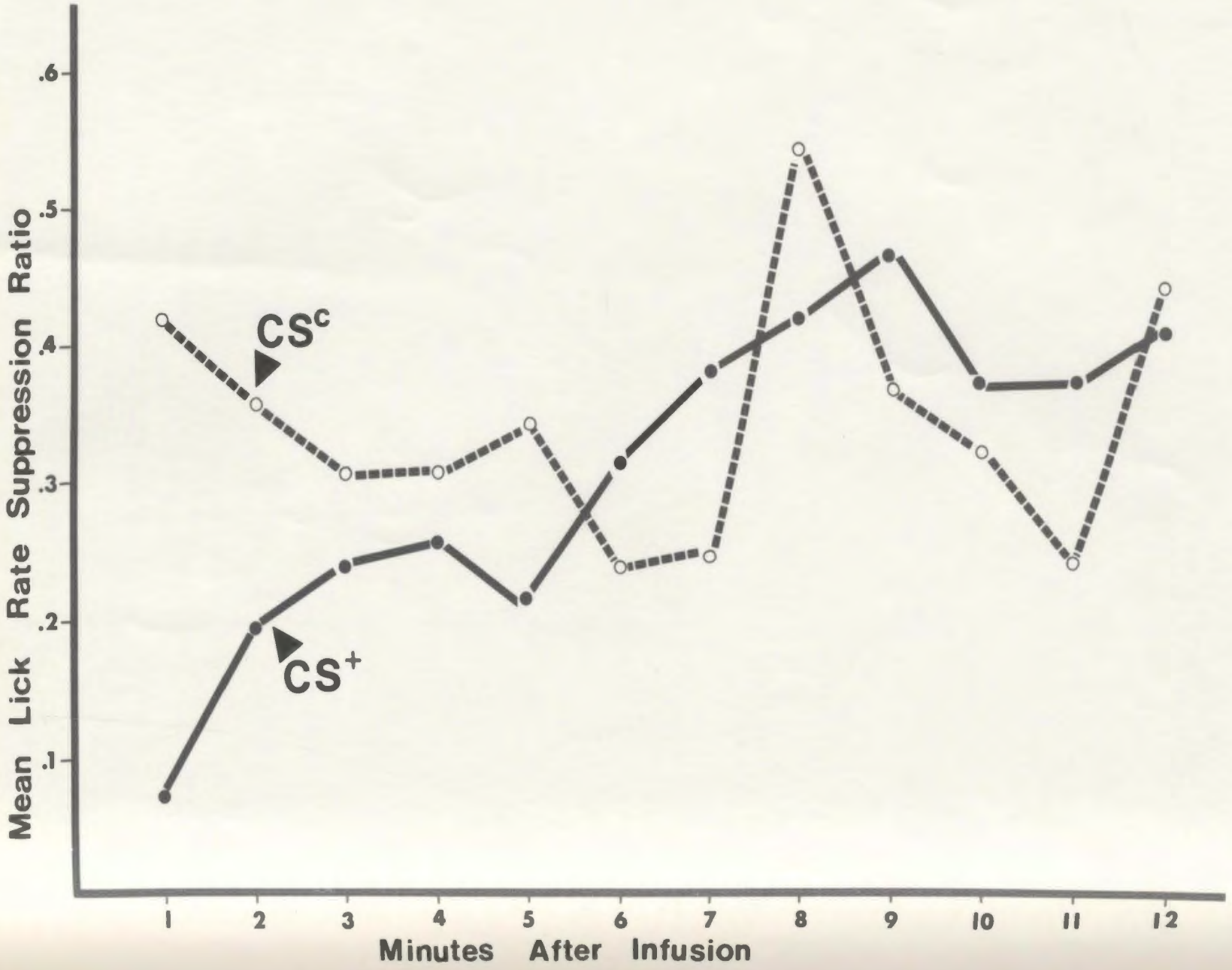

Mean Lick Rate Suppression Ratio
.6
.5
.4
.3
.2
.1
CS^C
CS^+
1
2
3
4
5
6
7
8
9
10
11
12
Minutes After Infusion

TABLE 4. Analysis of Variance Summary Table of the mean lick rate SRs in each block of three minutes in Experiment 2. A 2 x 2 x 4 unweighted means repeated measures ANOVA.

Source of variance	df	MS	F value	p
Between Subjects				
A (Infusion Condition)	1	13512.5	.123	
B (Flavor infused)	1	98531.9	.899	
AxB	1	29691.7	.271	
S	13	109593.0		
Within Subjects				
C (3 min.blocks)	3	73688.7	2.914	.05
AxC	3	58693.0	2.321	.10*
BxC	3	82614.9	3.266	.05
AxBxC	3	41047.9	1.623	
CxS	39	25291.7		

* Since the AC interaction was predicted apriori, it is significant as a one-tailed F ($p < .05$).

analyzed separately for a difference between CS+ and CSc conditions. The CS+ condition elicited greater suppression than the CSc condition in Minute 1 ($t(15)=5.80$; $p<.01$), but the conditions did not significantly differ in Minute 2 ($t(15)=1.47$; $p>.05$) or in Minute 3 ($t(15)= .13$). Of less theoretical interest, in the overall analysis, there was also a significant flavor of infusion x intervals interaction ($F_{(3,39)}=3.27$; $p<.05$); during the first three-minute block of licking, rats infused with sucrose had lower SRs than rats infused with NaCl ($t(15)=3.47$; $p<.01$).

It should be noted that since the clock which monitored the latency measure and the clock which monitored the lick rate measure both began immediately after the infusion of CS+ or CSc, the two measures of suppression were not independent until the rat completed the tenth lick. Therefore, the lower lick rate SRs seen in many of the CS+ exposed rats during the first minute actually reflected the fact that the rats had not returned to drink the TS; thus, the information provided by the lick rate measure was largely redundant.

The suppressed novel HCl intake by rats

infused with a lithium-paired CS+ flavor, suggests that the marginal difference between CS+ and CSc conditions in the five minute novel sucrose test of Experiment 1 was real. However, because the effect is short-lasting, it was not clearly visible when measured by the amount consumed in five minutes.

General Discussion

Obviously, the time to complete 10 licks was the most clearcut measure of the suppressive CR elicited by a flavor CS+. In fact, this measure was repeatedly shown to be the most sensitive indicator of conditioned suppression in the following series of experiments, when the time to begin drinking and the subsequent rate of licking were measured independently. In Experiments 3 - 6 which follow, immediately following an infusion of the CS+ or CSc flavor, the time to complete a criterion number of licks was measured; then after the rat reached the set criterion, the subsequent number of licks completed per 30 second interval was measured. Although in each experiment the CS+ and CSc conditions differed by the latency measure, in only one experiment did these con-

ditions differ by the subsequent lick rate measure. That is, in most cases, once the rats resumed drinking of the test solution, there was little further evidence of a suppressive CR. Therefore, in the following experiments, the measure which will be reported in the most detail will be that of latency to drink. Although the lick rate measure will also be reported, it will enter very little into the discussion of the findings.

Experiment 3

A new technique for measuring conditioned suppression of drinking following an exposure to a lithium-paired flavor CS+ was introduced in Experiment 2. By this measure, the CR elicited by a lithium-paired flavor CS+ suppressed the intake of a novel HCl solution for approximately one minute. Unflavored water was also shown to support a suppressive CR in Experiment 1^a, when the response measure was the overall amount consumed in three minutes. Therefore, Experiment 3 attempted to replicate the results of Experiment 1^a, with water as the test solution, using the technique devised in Experiment 2.

A second purpose of Experiment 3 was to determine the effect of stimulus intensity on the duration of the suppressive CR. Conceivably, a more intense CS+ flavor might result in a longer lasting suppressive CR than a less intense CS+ flavor. The concentration of the infused flavored solution was varied at the time of testing, but was constant among the groups at the time of conditioning; thus, Experiment 3 measured the

influence of CS intensity on the performance, rather than on the establishment, of a learned response.

The rats were given discrimination training between a sucrose solution and an HCl solution, both of a medium concentration. They were then tested in a manner similar to that of Experiment 2: They were presented a bottle of unflavored water TS and 30 seconds after they began to drink, half were exposed to a low concentration and half were exposed to a high concentration of their CS+ or CSc flavored solution.

Method

Subjects. The procedures were identical to those of the previous experiment except as indicated below. Forty male Sprague-Dawley rats ranging between 190-225 gms were implanted with intra-oral cannulae; however, during the course of the experiment, six animals were discarded because their cannula became dislodged. Throughout the experiment, the animals received their treatment in the morning when 18 hr. water deprived and were given access to water for 10 minutes, six hours later.

The rats were initially trained to drink from cups as described in Experiment 2. They were allowed 10 minute periods of access to water in a cup two times a day: The first was in the morning when the rats were 18 hr. deprived and the second was six hr. later.

Discrimination Conditioning (Days 4 - 7). The rats were then given discrimination training between 15% (w/v) Sucrose solution and 1.5% (v/v) HCl soluiton, counterbalanced for which was CS+ and which was CSc. The solutions were always presented in cups. As in Experiment 2, on Days 4 and 6, the rats were given 10 ml of the CS+ solution in a cup and 10 minutes later, when the cup was removed, were injected with 1.5 ml and 2.5 ml respectively of 2% LiCl. On Days 5 and 7, they received the same amount of the CSc solution as they had consumed of the CS+ solution on the preceding day, but were not injected after they drank. Six hours after each trial, the rats were presented 10 minutes of water in a cup.

Baseline Training and Suppression Test (Days 8 - 11). On each of two days following the final conditioning trial, the rats were given baseline training sessions in a manner similar to Experiment 2: The rats were presented a bottle of water,

and, thirty seconds after they began to drink, were infused with 2 ml of water over a 15 second period. Immediately after the infusion, the time to complete 10 licks was measured, and after the tenth lick, the number of licks completed in each 30-second interval was measured for 15 minutes. Therefore, unlike Experiment 2, the latency and the lick rate measures were obtained independently.

In the suppression test on Day 11, the rats were presented a bottle of unflavored water TS and, 30 seconds after they started to drink, were infused with 2 ml of either the CS+ flavor or the CSc flavor for 15 seconds. The concentration of the CS+ or CSc differed among the rats: 18 Ss were infused with the high concentration solution, 20% sucrose or 2% HCl, and 16 Ss were infused with the low concentration solution, 10% sucrose or 1% HCl. Thus, the final composition of the groups in the suppression test was as follows: High Sucrose CS+ (n=5), High HCl CS+ (n=4), Low Sucrose CS+ (n=4), Low HCl CS+ (n=4), High Sucrose CSc (n=5), High HCl CSc (n=4), Low Sucrose CSc (n=4), Low HCl CSc (n=4).

Design and Data Analysis. The design of Experiment 3 was a 2 x 2 x 2 factorial with the factors of Infusion condition, concentration of infused solution and flavor of infused solution. The latency scores were transformed into latency suppression ratios (SRs) as in Experiment 2 and were analyzed in a 2 x 2 x 2 unweighted means ANOVA.

Results

Latency Measure. Figure 5 presents the mean latency SR for each group in Experiment 3, pooled across the flavor infused. The groups were collapsed across the flavor of infusion because this factor did not effect the CR ($F_{(1,26)} = .013$). The two bars to the left represent the high (closed bar) and the low (open bar) concentrations of the infused CS+ flavor and the two bars to the right represent the high and low concentrations of the infused CSc flavor. The CS+ condition resulted in greater hesitancy to resume drinking of unflavored water than did the CSc condition ($F_{(1,26)} = 17.71$; $p < .001$) which replicated the results of Experiment 1[a]. However, as is seen in Table 5, no other effects were significant. The more intense CS+ flavor did not result in a more prolonged CR than did the less intense CS+ flavor;

Figure 5. Mean latency SR for groups in Experiment 3 when the TS was unflavored water. The closed bars represent the high concentration infused solution and the open bars represent the low concentration infused solution.

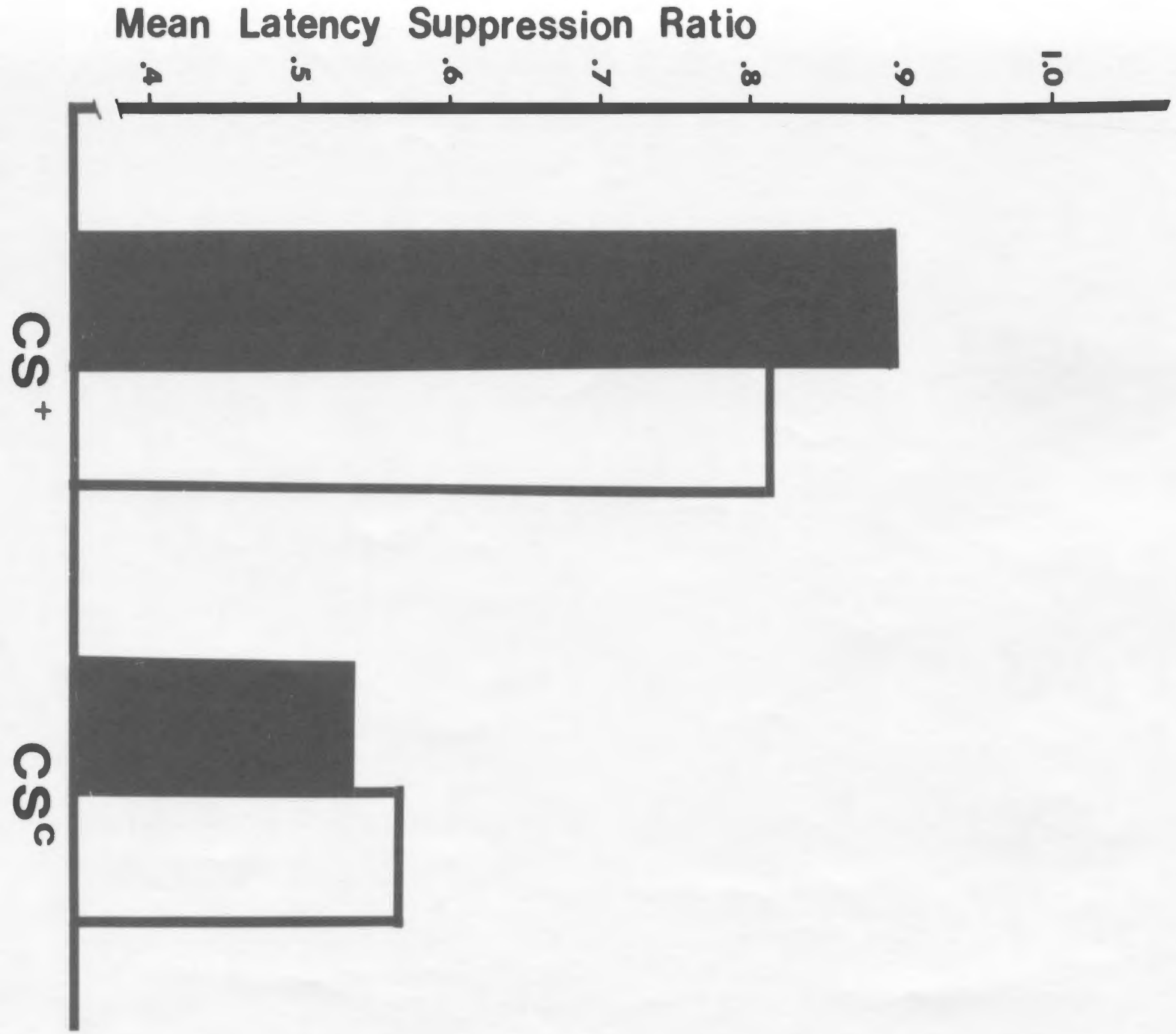

Mean Latency Suppression Ratio
.4
.5
.6
.7
.8
.9
1.0
CS+
CSc

TABLE 5. ANOVA Summary Table of latency SRs in Experiment 3.

Source of Variance	df	MS	F	p
A (Infusion Condition)	1	778111.630	17.754	.001
B (Concentration of infusion)	1	4358.414	.009	
C (Flavor of infusion)	1	573.163	.013	
AXB	1	25566.125	.583	
AXC	1	15678.750	.358	
BXC	1	90863.188	2.073	
AXBXC	1	756.250	.027	
S	26	43828.461		

in fact, the concentration of the infused solution did not influence the rats tendency to drink whether it was CS+ or CSc. Although stimulus intensity did not appear to effect the performance of the CR during testing, it may effect the acquisition of the CR during conditioning. This problem is addressed in Experiment 7.

Lick Rate Measure. The mean number of licks per minute after completion of the first 10 licks is presented for the various groups in Figure 6. A 2 x 2 x 5 unweighted means repeated measures ANOVA, presented in Table 6[a], was computed for the total number of licks completed in each 3 minute period of the test. There was no difference between the pattern of licking by the CS+ exposed rats and the CSc exposed rats ($F_{(1,26)} = 0.03$) which suggests that the CS+ elicited suppression was not noticeably maintained beyond the first 10 licks. The concentration of the infused solution, however, did effect the pattern of licking by both the CS+ and CSc groups ($F_{(1,26)} = 4.55$; $p < .05$). Those animals exposed to a highly concentrated solution showed greater suppression than those exposed to a weakly concentrated solution, but this effect did not vary

Figure 6. Mean number of licks of unflavored water TS completed, after the first 10 licks, by rats infused with either a high or low concentration of their CS+ or CSc flavored solution.

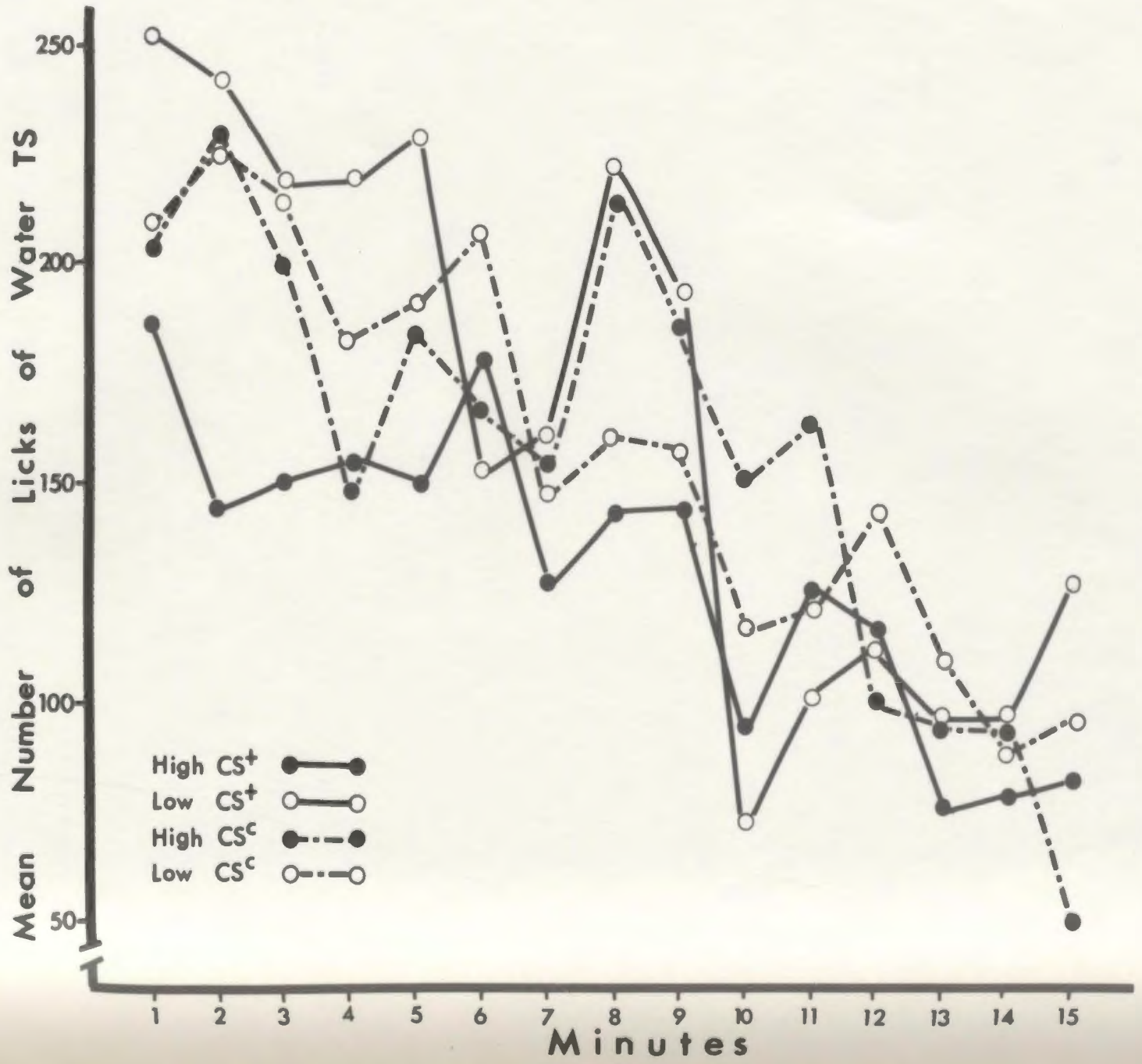

Mean Number of Licks of Water TS
250
200
150
100
50
High CS+
Low CS+
High CSc
Low CSc
1
2
3
4
5
6
7
8
9
10
11
12
13
14
15
Minutes

TABLE 6. ANOVA Summary Table for the lick rate data in Experiment 3.

6a. A 2 x 2 x 5 unweighted means repeated measures ANOVA for the entire test period, with the following factors: Infusion condition, Concentration of the infused solution and Blocks of licking (total number of licks in each 3 minute block of licking).

Source of Variance	df	MS	F	p
Between Subjects				
A (Infusion Condition)	1	467.788	.006	
B (Concentration)	1	330667.000	4.551	.05
AB	1	176525.000	2.429	
S	30	72662.300		
Within Subjects				
D (Blocks of licking)	4	603961.000	20.746	.001
AD	4	11592.000	.399	
BD	4	21520.500	.739	
ABD	4	14737.500	.506	
DS	120	29112.100		

6b. A 2 x 2 x 3 unweighted means repeated measures ANOVA for each of the first three minutes of licking.

Source of Variance	df	MS	F	p
Between Subjects				
A (Infusion Condition)	1	8424.773	.999	
B (Concentration)	1	53506.219	6.350	.025
AB	1	35011.008	4.155	.10
S	30	8426.481		
Within Subjects				
D (Minutes)	2	2240.197	.538	
AD	2	2746.088	.659	
BD	2	39.646	.010	
ABD	2	2013.750	.484	
DS	60	4164.270		

by Infusion Condition ($F_{(1,26)}$= 2.43). Finally, as is evident, the overall rate of licking decreased across the five three-minute blocks of licking ($F_{(4,104)}$= 20.75; $p < .001$).

Since the greatest differences between conditions were expected apriori early in the test period, an additional analysis tested for differences during the first three minutes of licking. A 2 x 2 x 3 unweighted means repeated measures ANOVA is presented in Table 6[b] for the number of licks in each of the first three minutes. The analysis indicated that the infusion condition x concentration interaction approached significance ($F_{(1,30)}$=4.15; $p < .10$). This suggested that Group High CS+ showed the greatest suppression during the first 3 minute period, but Newman-Keuls tests between the conditions revealed no differences. In addition, the rats exposed to high concentration solutions licked less during the first three minutes than those exposed to the low concentration solutions ($F_{(1,30)}$= 6.35; $p < .025$).

Discussion

As in the previous experiment, exposure to

the lithium-paired flavor CS+ suppressed rats tendency to resume drinking; however, the strength of the CS+ taste did not influence the degree of suppression. On the other hand, once the rats began to drink, the conditioned properties of the flavors no longer influenced the consumption of unflavored water TS, but the strength of the infused solution influenced the overall amount consumed. Once the rats began to drink, the strong tasting CSc produced suppression equivalent to that of the strong tasting CS+.

CHAPTER IV: TYPE OF TEST SOLUTION: EFFECT ON APPARENT MAGNITUDE OF SUPPRESSION

Experiments 4, 5 and 6 were concerned with the effects of the test solution on the strength of suppression elicited by an infusion of the CS+ flavor. That is, would the strength of the effect depend upon whether the rats consumed unflavored water, a novel flavored solution or a conditioned aversive flavored solution?

The traditional conditioned suppression literature suggests that, indeed, the type of test solution consumed would determine the strength of CS+ elicited suppression. The degree to which a fear eliciting CS suppresses ongoing responding is inversely related to the strength of the ongoing responding; ongoing behaviors of great strength may mask differences in the amount of fear elicited by the CS+ and, conversely, ongoing behaviors of very weak strength may be completely suppressed by any amount of fear (McAllister and McAllister, 1971). In particular, Vogel and Spear (1966) showed that the presentation of a CS previously paired with shock will suppress consumption of a 4% (w/v) Sucrose

solution to a greater extent than it will suppress consumption of a more highly preferred 32% (w/v) Sucrose solution which the rats normally drink at a faster rate.

The three experiments which follow were designed to map the duration of the suppressive CR elicited by a lithium-paired flavor CS+ when rats consume one of three types of Test Solutions: Unflavored water (Experiment 4), a novel flavored solution (Experiment 5) and a conditioned aversive flavored solution (Experiment 6). Each type of test solution supports a different rate of baseline drinking. A highly familiar unflavored water is readily consumed at a fast rate of licking. A novel flavored solution is consumed at a slower rate than is unflavored water, because rats are hesitant to consume substances which they have never previously tasted. Finally, a conditioned aversive flavored solution supports the weakest rate of drinking because rats consume very little of a flavored solution which predicts illness. Since each of these types of test solution supports a different rate of baseline drinking, they should support CS+ elicited suppressive CRs of different durations.

Experiments 4, 5 and 6 differed only on the basis of the type of test solution which the rats consumed; Experiments 5 and 6 were conducted at the same time and Experiment 4 began one week earlier. The first question to be answered by each experiment was: What is the duration of the suppressive CR that is supported by the given test solution? Then, once it was established that each type of test solution did support a suppressive CR, the CR durations were compared across Experiments 4 - 6.

Since the procedures of Experiments 4, 5 and 6 differed from those of the previous experiments, I have outlined each phase below.

Phase 1 Conditioning. This conditioning phase served to establish a conditioned aversive test solution (TS). Although the Phase 1 conditioned aversive solution was only used during testing in Experiment 6, in order to maintain constant pre-testing treatments in Experiments 4, 5 and 6, all rats received this initial conditioning trial. The rats were presented a cup containing 10 ml of a novel flavored solution, 1.25% (w/v) Coffee or 1.25% (w/v) NaCl, for 10 minutes and were then injected with 1 ml of 2% (w/v) LiCl. The 1 ml

dose was selected in order to establish a reliable aversion of a moderate strength.

Discrimination Conditioning. This phase served to establish the discriminative CS+ and CSc solutions. The procedures were similar to those of Experiment 2 and 3.

Baseline Training. On each of five days, the rats were given baseline training trials. These five trials established a stable baseline licking response prior to the suppression test. A stable response was important because the results were reported as raw latency scores which designated the actual duration of the suppressive CR, rather than as the suppression ratios, used in the previous experiments, which indicated merely the presence or absence of a CR.

On each baseline day, the rats were allowed to consume unflavored water from a bottle for 30 seconds before they were infused for 15 seconds with 2 ml of unflavored water. Immediately following the infusion, the drinkometer system was activated to measure the immediate aftereffect of the infusion.

One measure of the immediate aftereffect was the latency to complete the first two licks. The

criterion was changed from the 10 lick criterion of Experiments 2 and 3 because I wanted a measure of the time to resume licking which could be compared across Experiments 4, 5 and 6. In Experiment 6, the rats were to be tested with a conditioned aversive TS. Since their rate of licking was expected to be low, it was conceivable that neither the rats exposed to the CS+ nor those exposed to the CSc would reach the 10 lick criterion within a pre-determined maximum period of 10 minutes, even though they had returned to sample the Aversive TS. Therefore, the two lick criterion was a more realistic response requirement for the measure of latency to resume drinking than was the 10 lick criterion.

Once the rats completed the second lick, the number of licks completed within each 30 second interval was measured for three minutes. This lick rate measure was expected to detect the residual suppression elicited by the CS+ infusion after the rats began to drink.

Suppression Tests. The procedures of the suppression tests were identical to those of the baseline training trials, except that the solutions infused and the solutions consumed differed. The

rats were presented a bottle containing the appropriate test solution depending upon the experiment and, 30 seconds after they began to drink, were infused with the discriminative CS+ or CSc flavored solution. The CR measures were: Latency to lick 2 licks, thirty-second lick rate for three minutes and the total amount of TS consumed (on Test Day 1 only). The same procedure was employed on each subsequent day until the suppressive CR had extinguished.

Avoidance Test. After the suppressive CR had extinguished, the rats were given single bottle preference tests with their discriminative CS+ and CSc solution. This phase determined whether or not attenuation of the suppressive CR would also result in attenuation of the avoidance response to the CS+ flavored solution.

Experiment 4

Experiment 4 was conducted to measure the duration of the CS+ elicited suppressive CR when rats consumed an unflavored water TS.

Method

Subjects. Twenty-four male Sprague-Dawley rats between 225 and 250 gms were implanted with intraoral cannulae as described in Experiment 2. For the following three days, they were allowed free access to water. After the recovery period, the rats were maintained on an 18 hr water deprivation schedule throughout the remaining experimental manipulations; six hr after each treatment session, they were given access to water for an additional ten minutes. On the first three post-recovery days (Days 1 - 3), the rats were trained to consume their daily water from cups.

Phase 1 Conditioning. On Day 4, all rats were given the Phase 1 conditioning trial in which either Coffee or NaCl was made aversive, as described in the introduction. This phase was included to equate the rats in Experiments 4 - 6 for previous toxicosis conditioning experience;

however, the phase 1 conditioned aversive solution was not used in the testing phase of the present experiment. On the following day (Day 5), the rats received water in a cup for 10 minutes.

Discrimination Conditioning Trials. On Days 6 - 9, the rats were given Discrimination Conditioning trials as in Experiment 2. The discriminative flavors were 20% (w/v) Sucrose solution and 1.5% (v/v) HCl solution. On Days 6 and 8, the rats received a CS+ training trial: They were presented 10 ml of their CS+ flavored solution in a cup for 10 min. and were then injected with 1.5 ml and 2.5 ml respectively of 2% (w/v) LiCl. On Days 7 and 9, the rats received a CSc training trial: They were presented the same amount of their CSc flavored solution as they had consumed of their CS+ flavored solution on the previous day, but the rats were not injected after they drank.

Baseline Training Trials. On the following five days (Days 10 - 14), the rats were given baseline training trials. As in the previous experiments, the food was removed from each rats cage 30 min. prior to a trial and was replaced 30 min. after a trial. In each session, the rat was

presented a bottle; thirty seconds after it began to drink, the rat was given a 2 ml intraoral infusion of water over a 15 second period. The drinkometer system was activated immediately following the infusion to measure the latency to complete two licks and the subsequent number of licks per 30 seconds for three minutes. On the final baseline training day, each rat had completed two licks of water within 10 seconds after an infusion of water.

Suppression Tests. On Day 15, the rats were tested for conditioned suppression of unflavored water consumption. Thirty seconds after the rats began drinking from a bottle containing the water TS, they were infused with 2 ml of either the discriminative CS+ or CSc flavored solution over a 15 second period. Immediately following the infusion, the latency to complete two licks of water, the subsequent lick rate per 30 second interval and the total amount of water TS consumed (only on Test Day 1) were measured. The identical procedure was continued on Days 16 and 17 until the suppressive CR had extinguished.

Avoidance Test. All rats were given a single bottle preference test for their discriminative CS+

and CSc flavored solutions after the suppressive CR had extinguished. These tests determined whether or not extinction of the suppressive CR weakened the aversion to the discriminative CS+ flavored solution. On Days 18, 19, 20 and 22, the rats received three minutes of water during the treatment session. On Day 21, the rats were given three minutes of access to a single bottle which contained either the discriminative CS+ or CSc flavored solution and on Day 23, they were presented the alternate discriminative conditioned solution; the presentation order was counterbalanced on the basis of prior history during the suppression tests.

Design. The design of Experiment 4 was a 2 x 2 factorial with six subjects per group. The factors were: Infusion condition and flavor of the infused solution.

Results and Discussion

Suppression Tests. The mean latency to complete two licks of unflavored water TS after a brief exposure to CS+ or CSc is presented in Figure 7 for each test day. It was necessary to transform the Day 1 raw latency scores into square roots, because the

Figure 7. Mean latency to complete 2 licks of unflavored water TS on each test day of Experiment 4.

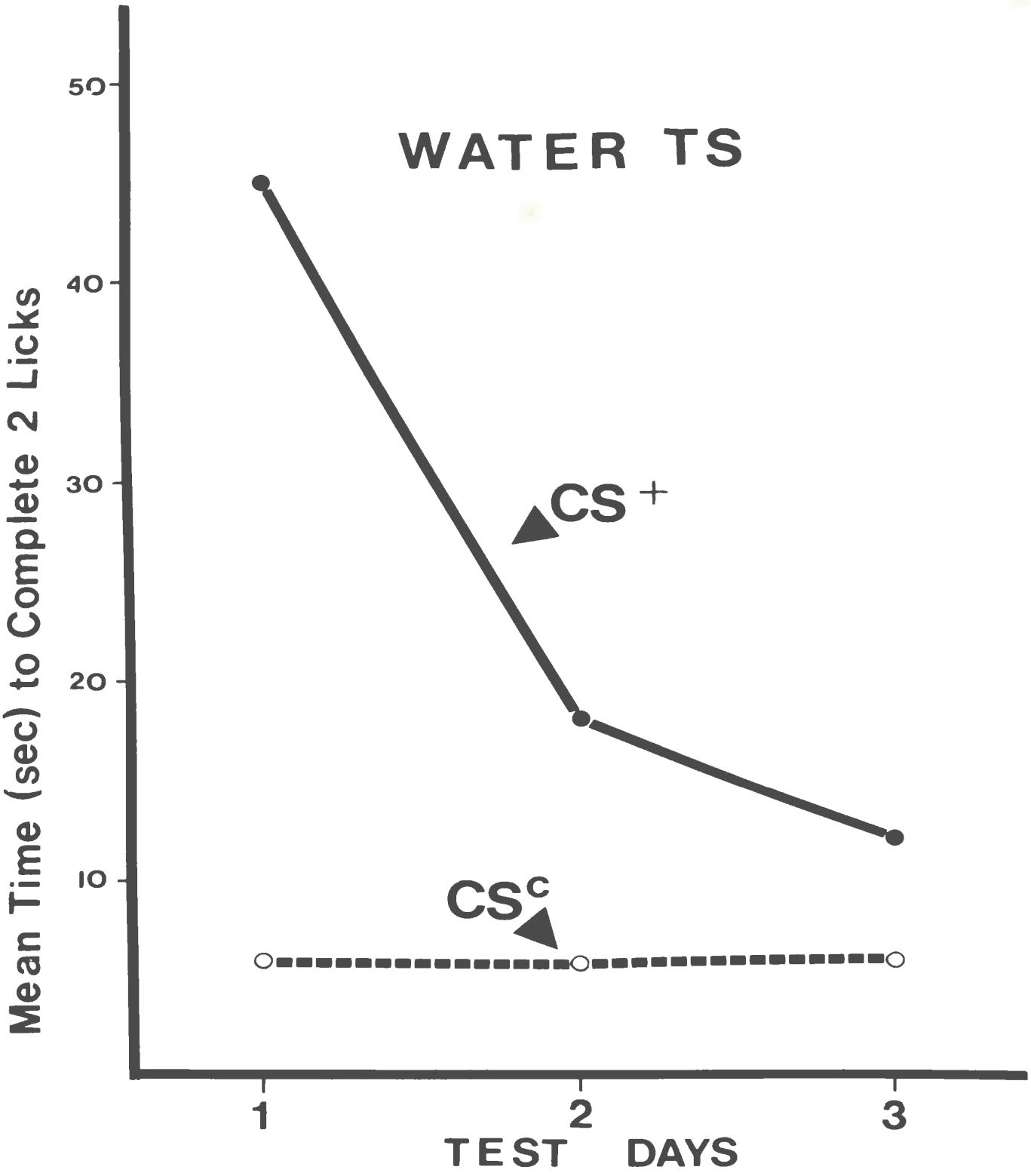

WATER TS
Mean Time (sec) to Complete 2 Licks
50
40
30
20
10
CS +
CS C
1
2
3
TEST DAYS

TABLE 7. ANOVA Summary Table for the square root latency scores on Test Day 1 of Experiment 4 with unflavored water TS.

Source of Variance	df	MS	F value	p
A (Infusion Condition)	1	153.348	78.171	.001
B (Flavor of Infusion)	1	.019	.009	
AB	1	.335	.166	
S	20	2.013		

within group variability was greater in the CS+ group than in the CSc group ($F_{max}(2,11)= 4.01$; $p<.05$). The 2 x 2 ANOVA for the square root of the raw latency scores on Test Day 1 is presented in Table 7 and the results of the analyses for the remaining days are presented in Appendix C. On Test Day 1, rats infused with CSc resumed drinking unflavored water within a mean of 6 seconds, but rats infused with CS+ hesitated for a mean of 45 seconds ($F_{(1,20)}= 76.17$; $p<.001$). This difference was maintained on Test Day 2 ($F_{(1,20)}= 7.82$; $p<.01$), but was no longer significant on Test Day 3 ($F_{(1,20)}= 3.21$; $p>.05$). The flavor of the infused solution did not influence the rats hesitancy to resume drinking on any test day.

Figure 8 presents the mean number of licks completed within each 30-second interval on Test Day 1 by Group CS+ and Group CSc. Although Figure 8 suggests that Group CS+ licked at a faster rate overall than Group CSc, the 2 x 6 repeated measures ANOVA presented in Table 8 revealed no significant differences between groups. Furthermore, a comparison of the trend of Groups CS+ and CSc on Test Day 1 also revealed no significant differences ($F_{(1,100)}=2.33$).

Figure 8. Mean number of licks completed per 30 second interval of unflavored water TS by CS+ and CSc infused rats on Test Day 1 of Experiment 4.

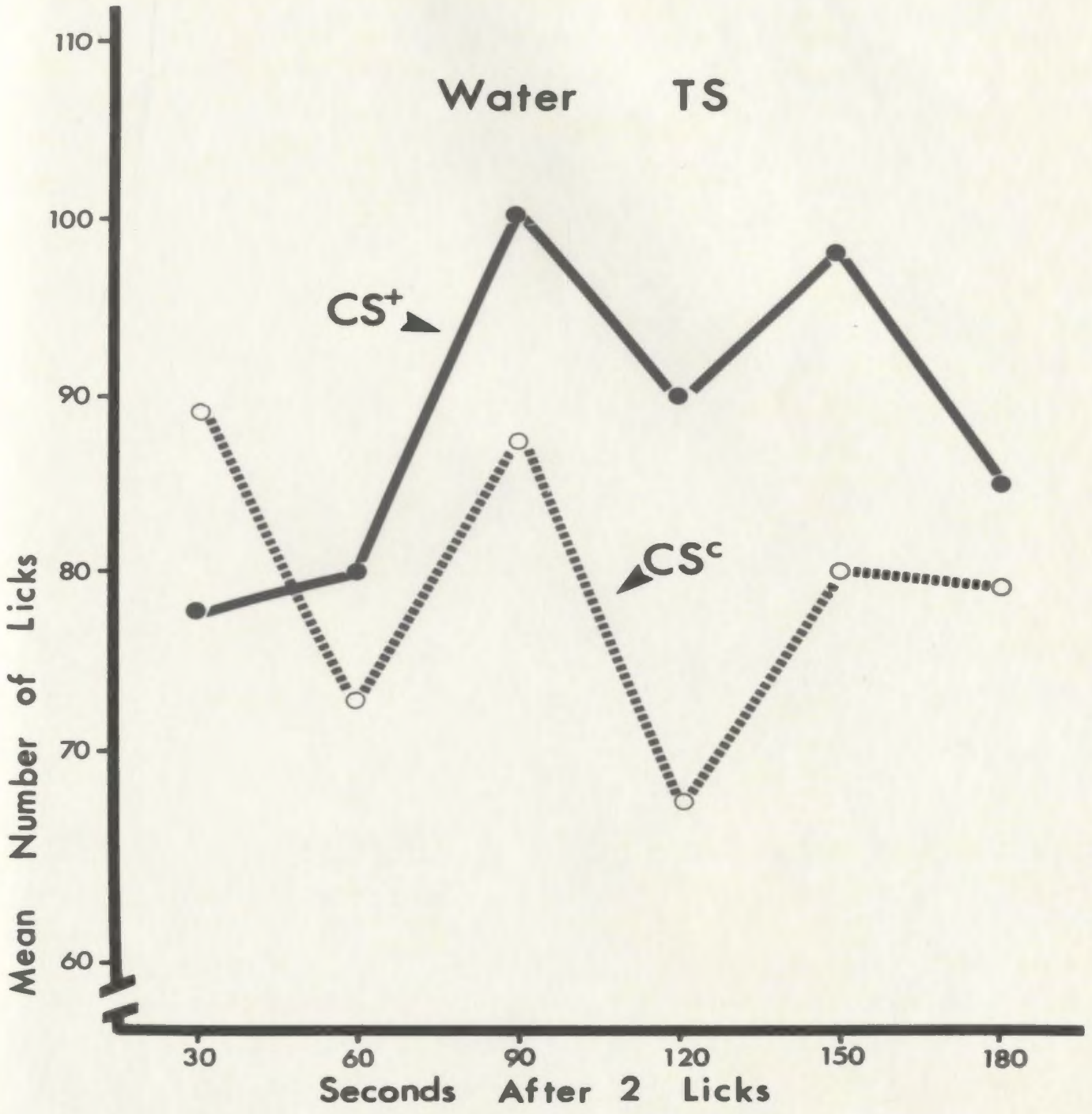

Water TS
110
100
90
80
70
60
CS+
CSc
Mean Number of Licks
30
60
90
120
150
180
Seconds After 2 Licks

TABLE 8. ANOVA Summary Table for the lick rate data on Day 1 of Experiment 4 with water TS. A 2 x 6 repeated measures ANOVA included the factors of Infusion condition and 30-second intervals.

Source of Variance	df	MS	F value
Between Subjects			
A (Infusion Condition)	1	1122.700	.47
S	22	2408.200	
Within Subjects			
B (30-sec. intervals)	5	1076.240	1.34
AB	5	1170.480	1.46
SB	110	802.440	

The groups did not differ on any of the remaining test days as shown in Appendix C. Therefore, although the rats were more hesitant to resume drinking an unflavored water test solution after a brief exposure to the CS+ flavor, they showed no further suppression once they had completed two licks.

Finally, the suppressive CR was not evident by the amount of water TS consumed on Suppression Test Day 1 ($F_{(1,22)} = 1.64$); Group CS+ drank a mean of 5.7 ml and Group CSc drank a mean of 6.1 ml. This result suggests that the CS+ elicited suppression of unflavored water consumption in Experiment 1a was largely the result of the rats' hesitancy to resume drinking.

Avoidance Test. When tested for amount consumed of the discriminative flavored solutions, the rats drank more of the CSc flavored solution (M=9.0 ml) than of the CS+ flavored solution (M= 1.4 ml) with $F_{(1,46)} = 98.28$ ($p < .001$). This aversion was not effected by the three prior extinction exposures to the CS+ flavor in Group CS+. There was no difference in the amount of CS+ flavored solution consumed by the rats infused with the CS+ during the suppression test and those infused with the CSc during the suppression test ($F_{(1,22)} < 1.0$).

Experiment 5

In Experiment 4, rats suppressed their drinking of an unflavored water TS for 45 seconds following an exposure to a lithium-paired flavor CS+; however, the CR was no longer apparent once the rats returned to drink the TS. A more prolonged CR might be elicited by the CS+ flavor when the rats are tested with a solution which is less readily consumed than unflavored water. In Experiment 5, the TS was a novel flavored solution which rats are more hesitant to drink than unflavored water.

Method

Twenty-four male Sprague-Dawley rats between 220 - 248 gms were treated identically as those in Experiment 4 except as specified below.

On Day 15, the rats were given a suppression test with a novel flavored test solution. Half of the rats were presented a novel 1.25% (w/v) Coffee solution and the other half were presented a novel 1.25% (w/v) NaCl solution. Thirty seconds after a rat began to drink the novel TS, it was infused with 2 ml of the CS+ or CSc solution from discrimination training; the latency to complete two

licks, the number of licks per 30 second interval following the first two licks and the amount consumed in the test period were measured. The same procedure was followed on Days 16 - 19.

In the avoidance tests on Days 22 and 24, the rats were given a single bottle presentation of their discriminative CS+ and CSc flavored solutions as described in Experiment 4. In addition, on Day 26, the rats were given a three - minute single bottle test with their TS from the suppression tests; this determined whether the previous pairing of CS+ or CSc with the TS influenced the preference for the TS. On all intervening days, Days 20, 21, 23 and 25, the rats were given water during the three-minute treatment period.

The design of Experiment 5 was a 2 x 2 x 2 factorial with three rats per condition. The factor of theoretical interest was the infusion condition and the two additional counterbalancing control factors were Flavor of Infusion (Sucrose or HCl) and Flavor of TS (Coffee or NaCl).

Results and Discussion

Suppression Tests. Figure 9 presents the mean latency to complete 2 licks of the Novel TS

Figure 9. Mean latency to complete 2 licks of a novel flavored TS on each test day of Experiment 5.

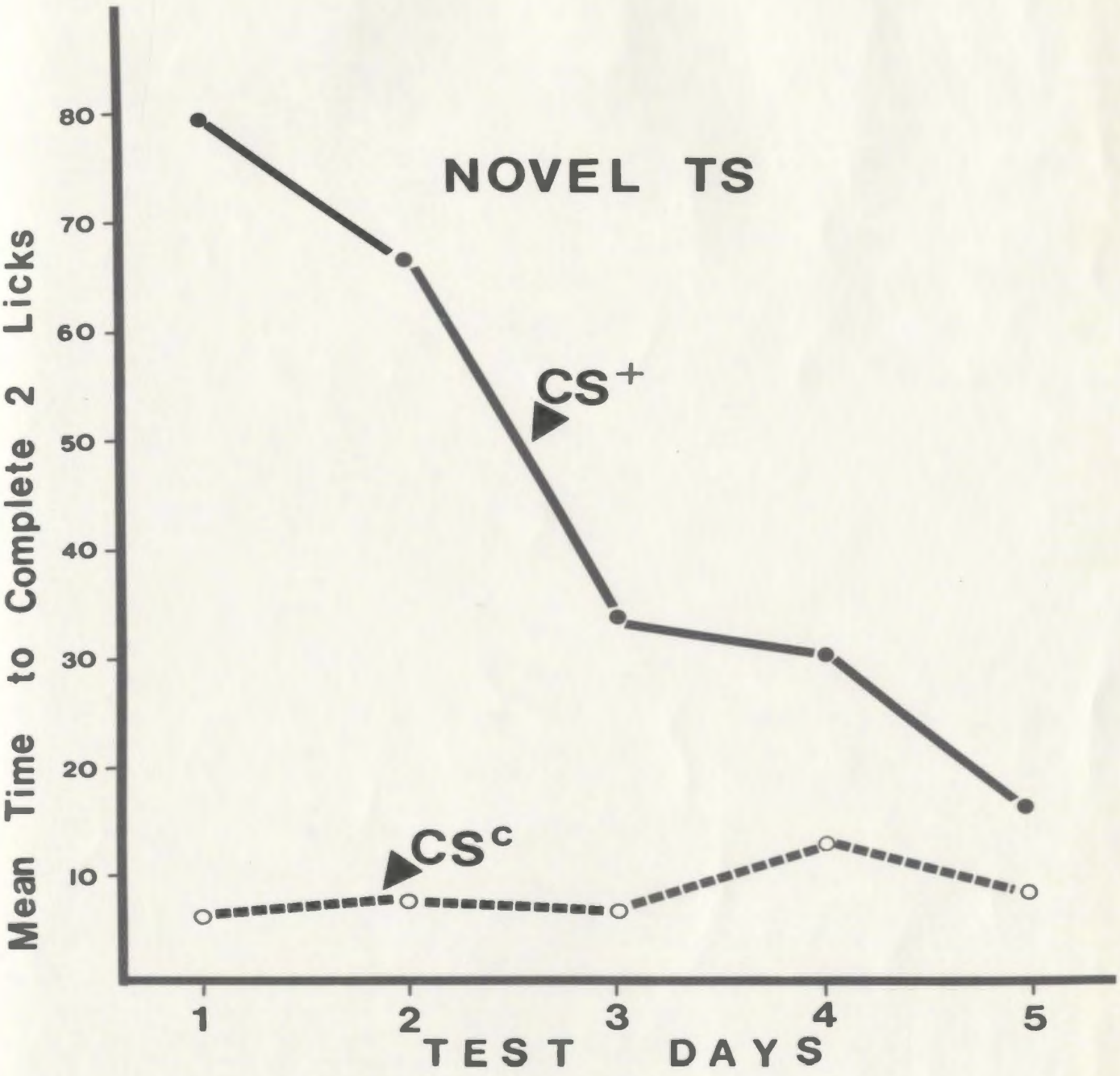

NOVEL TS
Mean Time to Complete 2 Licks
80
70
60
50
40
30
20
10
CS+
CSC
1
2
3
4
5
TEST DAYS

TABLE 9. ANOVA Summary Table for the square root latency scores on Test Day 1 of Experiment 5 with a novel TS.

Source of Variance	df	MS	F value	p
A (Infusion Condition)	1	229.952	38.732	.001
B (Flavor of Infusion)	1	1.730	.291	
C (Flavor of TS)	1	.935	.157	
AB	1	2.542	.428	
AC	1	4.748	.800	
BC	1	2.381	.401	
ABC	1	3.077	.518	
S	16	5.937		

by the CS+ and CSc exposed groups on each test day. The results of the 2 x 2 x 2 ANOVA for the square root of the latency scores on Test Day 1 are presented in Table 9 and the results of the analyses for the remaining test days are presented in Appendix C. Neither the flavor of the CS nor the flavor of the TS influenced the latency to resume drinking. In fact, the only significant effect on any test day was that of Infusion Condition. As is evident from Figure 9, the greatest difference between the CS+ and CSc conditions occurred on Test Day 1 ($F_{(1,16)} = 38.73$; $p<.001$). The rats infused with CS+ had a mean latency of 80 seconds, while the rats infused with CSc resumed drinking within a mean of 6 seconds. This suppressive effect gradually weakened across the test days, but remained significant ($F's_{(1,16)}>8.77$; $p's<.01$) until Day 5 ($F_{(1,16)}= 4.48$; $p>.05$).

The mean number of licks of novel flavored TS per 30 second interval for Groups CS+ and CSc on Test Day 1 are presented in Figure 10. These results were analyzed in a 2 x 6 repeated measures ANOVA as presented in Table 10. Group CS+ showed greater overall suppression of licking than Group CSc ($F_{(1,22)}= 10.04$; $p<.01$), and this difference varied across the 30-second intervals ($F_{(5,110)}= 3.14$;

Figure 10. Mean number of licks completed per 30-second interval of novel flavored TS on Test Day 1 by Groups CS+ and CSc in Experiment 5.

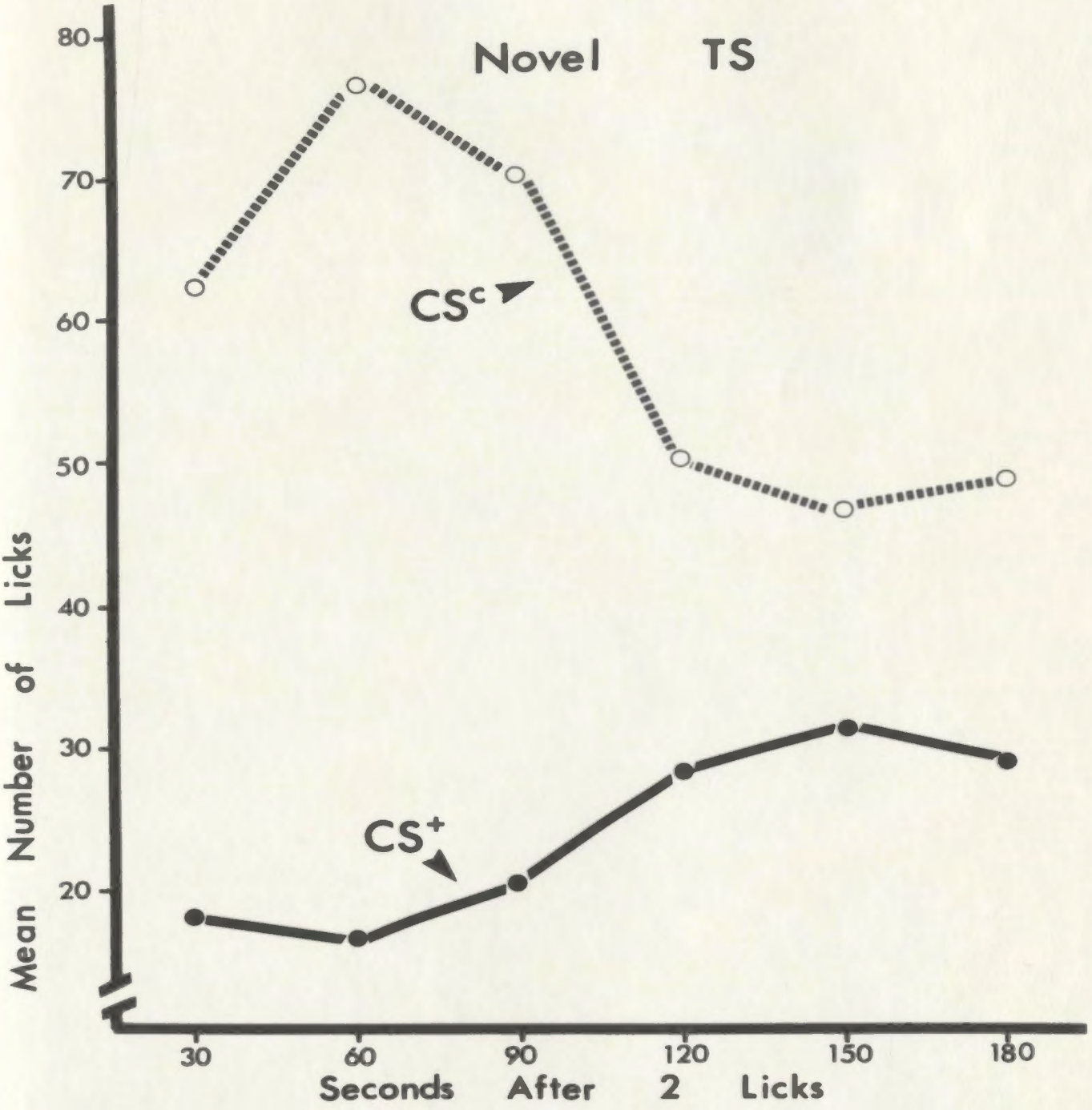

Novel TS
80
70
60
50
40
30
20
CS^c
CS^+
Mean Number of Licks
30
60
90
120
150
180
Seconds After 2 Licks

TABLE 10. ANOVA Summary Table for the lick rate data on Day 1 of Experiment 5 with a novel TS. The 2 x 6 repeated measures ANOVA included the factors of Infusion Condition and 30-second intervals.

Source of Variance	df	MS	F value	p
Between Subjects				
A (Infusion Condition)	1	50887.687	10.04	.01
S	22	5067.461		
Within Subjects				
B (30 sec. intervals)	5	279.204	.325	
AB	5	2696.496	3.143	.025
SB	110	857.968		

$p < .025$). Group CS+ licked at a slower rate during each of the first three 30 second intervals than Group CSc (t's (22) > 2.93; $p < .001$), but they did not differ in the last three intervals (t's (22) < 1.53; p's $> .05$). The groups did not differ on any other test day, as is shown in Appendix C.

Finally, on Test Day 1, the amount of novel flavored TS consumed by the CS+ (M=3.4 ml) and the CSc (M=4.7 ml) exposed groups did not significantly differ ($F_{(1,22)} = 2.38$; $p > .05$).

Avoidance Tests. In the subsequent avoidance tests with the discriminative CS flavors, the rats consumed more of the CSc solution (M= 9.6 ml) than of the CS+ solution (M= 1.6 ml) with $F_{(1,46)} = 97.71$ ($p < .01$). Also, as in Experiment 4, although the suppressive CR elicited by the CS+ exposures had weakened by the fifth test day, the aversion to the CS+ was not effected. The amount of the CS+ flavored solution consumed by the rats which had been previously infused with CS+ did not differ from the amount consumed by the rats which had been previously infused with CSc ($F_{(1,22)} < 1.0$).

Finally, rats which had been exposed to CS+ and rats which had been exposed to CSc drank similar amounts of the TS when tested on Day 26 ($F_{(1,22)} < 1.0$).

The contiguous presentation of the CS+ and the novel TS did not influence the rats' preference for the TS; apparently, the TS gained neither excitatory nor inhibitory properties.

Experiment 6

The rats took longer to resume drinking a novel flavored TS (approximately 80 seconds) than an unflavored water TS (approximately 45 seconds) immediately following an exposure to a lithium-paired flavor CS+. If the enhanced suppression with the novel TS was the result of a slower baseline drinking response, then a conditioned aversive TS should support an even greater suppressive CR than the novel TS. In Experiment 6, the rats were tested with the Phase 1 conditioned aversive solution.

Method

Twenty-four male Sprague-Dawley rats between 222 - 250 gms were treated exactly as the rats in Experiment 4 except as specified below.

The rats were given the first suppression test on Day 15. They were presented a bottle containing the Phase 1 conditioned aversive solution; for half of the rats this was 1.25% (w/v) Coffee and for the other half this was 1.25% (w/v) NaCl. Thirty seconds after the rats began to drink the TS, they were given a 2 ml, 15 second infusion of the discriminative CS+ or CSc flavor. The latency to

complete two licks, the subsequent lick rate per 30 seconds and the amount consumed (on Day 15 only) were measured. The same procedure was followed on Days 16 - 19.

On Days 22 and 24, the rats were given the single bottle avoidance test with the discriminative CS+ and CSc solutions as previously described in Experiment 4.

Results and Discussion

Suppression Tests. The mean latency to complete two licks of the conditioned aversive flavored TS by the CS+ and the CSc groups on each test day is presented in Figure 11. The 2 x 2 x 2 ANOVA for Test Day 1 is presented in Table 11. The only significant effect was that of Infusion condition ($F_{(1,16)} = 14.14$; $p < .01$); the rats infused with CS+ (M= 235 seconds) took longer to resume drinking than the rats infused with CSc (M= 62 seconds). The CS+ elicited suppressive effect was maintained on Test Day 2 ($F_{(1,16)} = 6.7$; $p < .025$), but had weakened by Test Day 3 ($F_{(1,16)} = 3.4$; $p > .05$) and was not evident on Days 4 and 5. The results of the 2 x 2 x 2 ANOVAs for each of Test Days 2 - 5 are presented in Appendix C.

Figure 11. Mean latency to complete 2 licks of a conditioned aversive TS on each test day of Experiment 6.

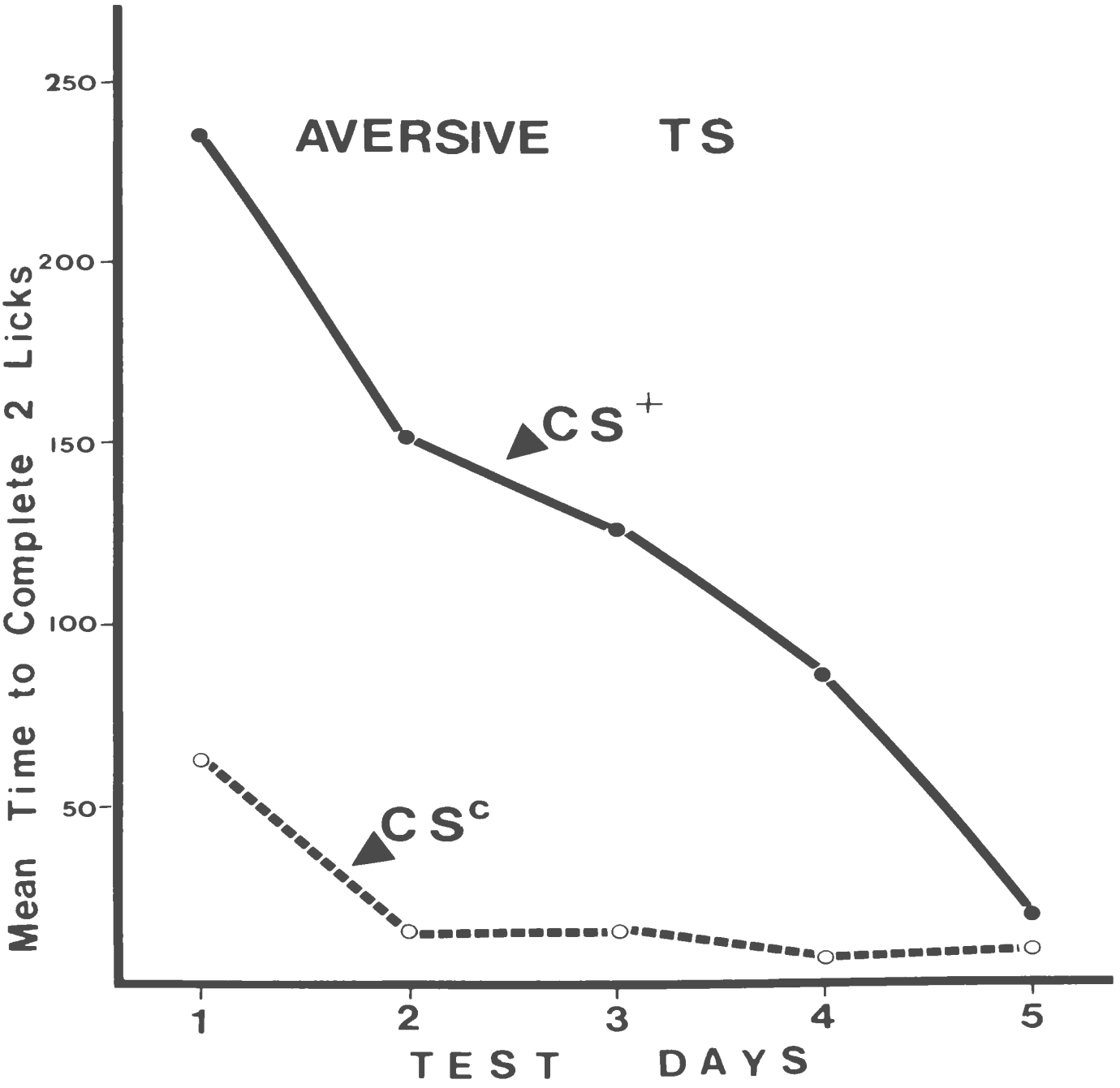

AVERSIVE TS
250
200
150
100
50
Mean Time to Complete 2 Licks
CS+
CSC
1
2
3
4
5
TEST DAYS

TABLE 11. ANOVA Summary Table for the square root latency scores on Test Day 1 of Experiment 6 with a conditioned aversive TS.

Source of Variance	df	MS	F value	p
A (Infusion Condition)	1	352.529	14.136	.01
B (Flavor of Infusion)	1	20.639	.828	
C (Flavor of TS)	1	1.688	.068	
AB	1	.591	.024	
AC	1	17.113	.686	
BC	1	1.179	.047	
ABC	1	10.776	.432	
S	16	24.938		

Figure 12 presents the mean number of licks of conditioned aversive TS per 30 second interval on Day 1 by groups infused with CS+ or CSc. The results of a 2 x 6 repeated measures ANOVA for Day 1, presented in Table 12, indicated that the only effect which approached significance was an infusion condition x intervals interaction ($F_{(5,110)}= 2.21$; $p<.10$). Since a difference in the pattern of licking was expected <u>apriori</u> to occur between Groups CS+ and CSc, the groups were compared by a subsequent trend analysis across the six 30-second intervals. There was, indeed, a difference in the pattern of drinking between Groups CS+ and CSc ($F_{(1,110)}= 4.24$; $p<.05$). During the first 30-second interval of licking, Group CS+ licked less than Group CSc ($t(22)= 1.90$; $p<.05$), but they did not differ during any other interval (t's (22) $<$ 1.07; p's $>$ .10). The groups did not differ on any other test day, as reported in Appendix C.

Finally, the CS+ exposed and the CSc exposed rats drank similar amounts of the aversive TS on Test Day 1 ($F_{(1,22)}= 1.32$).

<u>Avoidance Tests</u>. The rats showed an aversion to the discriminative CS+ (M= 1.6 ml) flavored solution when compared with the CSc (M=10.9 ml)

Figure 12. Mean number of licks per 30-second interval of a conditioned aversive TS on Suppression Test Day 1 by Groups CS+ and CSc in Experiment 6.

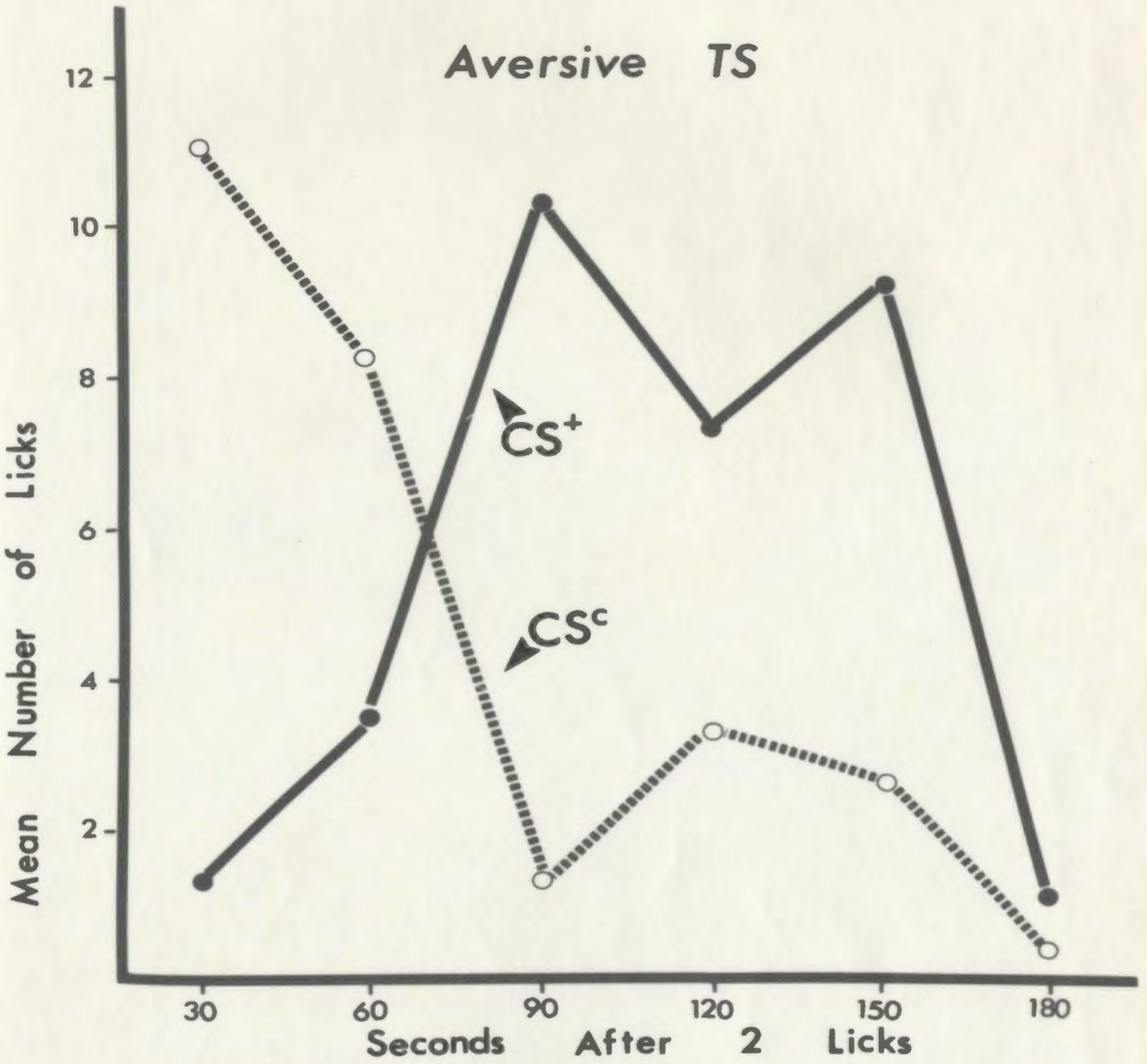

Aversive TS
12
10
8
6
4
2
Mean Number of Licks
CS+
CSc
30
60
90
120
150
180
Seconds After 2 Licks

TABLE 12. ANOVA Summary Table for the lick rate data on Day 1 of Experiment 6 with a conditioned aversive TS. The 2 x 6 repeated measures ANOVA included the factors of Infusion Condition and 30-second intervals.

Source of Variance	df	MS	F value	p
Between Subjects				
A (Infusion Condition)	1	21.777	.31	
S	22	691.770		
Within Subjects				
B (30 sec. intervals)	5	109.033	.74	
AB	5	323.927	2.21	.10
SB	110	146.656		

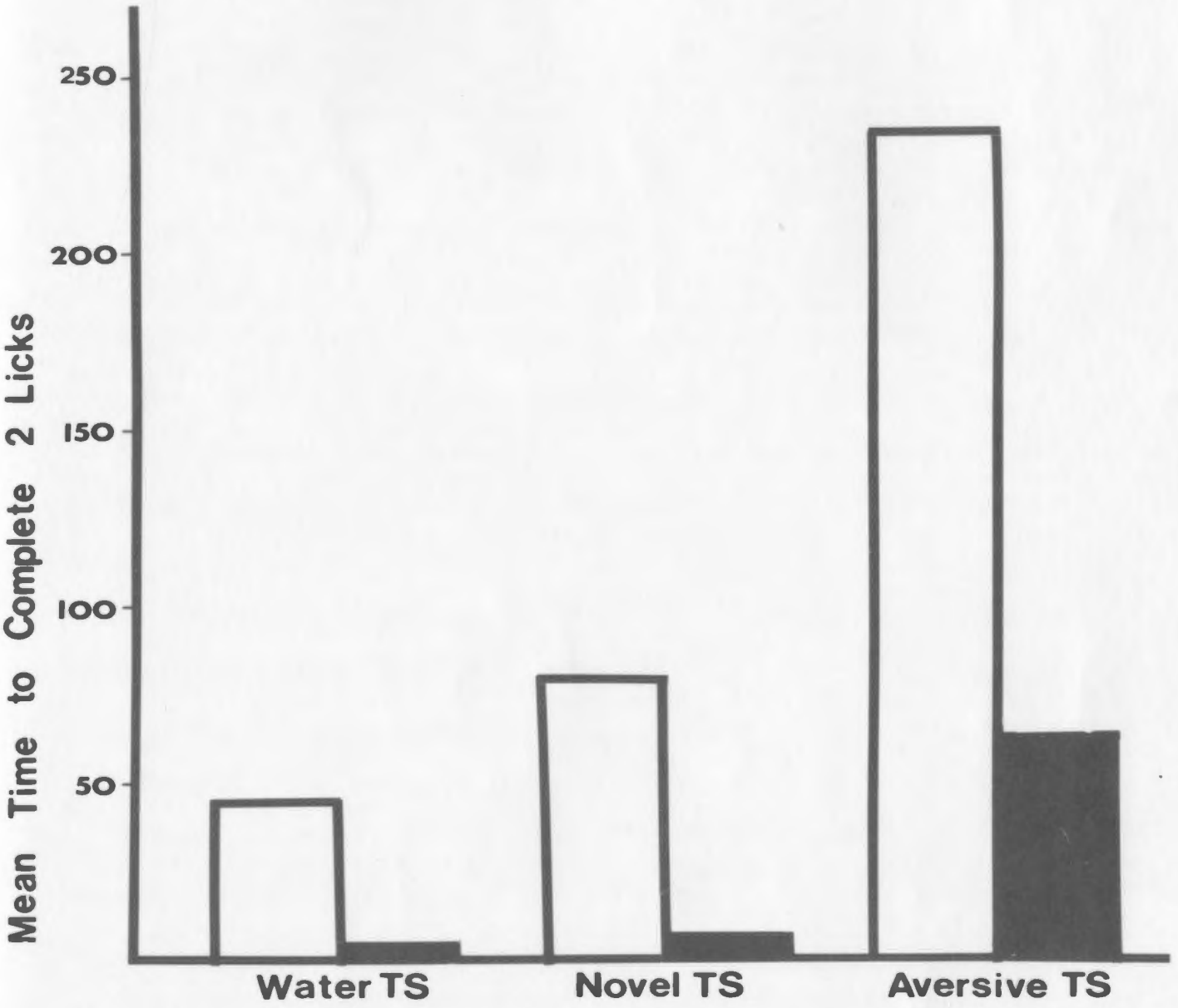
flavored solution ($F_{(1,46)} = 189.95$; $p < .001$). This aversion did not weaken after the five CS+ exposures during the suppression test, because the amount of discriminative CS+ flavored solution consumed did not differ between rats with a previous history with CS+ exposures and rats with a history with CSc exposures ($F_{(1,22)} < 1.0$). Thus, as in Experiments 4 and 5, although the suppressive CR elicited by the lithium-paired CS+ had weakened considerably by the final suppression test, the aversion to that flavor was not effected.

Comparison of Results Across Experiments 4, 5 and 6

The most sensitive measure of the suppressive CR in each of Experiments 4, 5 and 6 was the latency to complete two licks. When measured by the lick rate measure, only the Novel TS (in Experiment 5) clearly supported the suppressive CR, and this was only evident on Test Day 1. Therefore, to determine whether the preference for the test solution determined the strength of the CS+ elicited suppressive CR, only the latency results will be used.

Figure 13 presents the mean time to complete 2 licks of each type of TS by the rats exposed to either the lithium-paired CS+ flavor or the equally familiar, but safe, CSc flavor. Earlier studies with a shock-paired CS+ (e.g., Vogel and Spear, 1966) suggest that the rat's baseline preference for the TS would determine the strength of the suppression elicited by the CS+. Therefore, it was first necessary to demonstrate that the three types of test solutions did, in fact, support differing tendencies to drink. A 2 x 3 ANOVA was performed on the TS intakes on Suppression Test Day 1 of the

Figure 13. Mean time to complete 2 licks of TS in each of Experiments 4 (Water TS), 5 (Novel TS) and 6 (Aversive TS) by rats exposed to either CS+ (open bars) or CSc (closed bars).


Mean Time to Complete 2 Licks
250
200
150
100
50
Water TS
Novel TS
Aversive TS

CS+ or CSc groups in each experiment. Indeed, the overall amount consumed of the various test solutions differed significantly ($F_{(2,66)}$= 33.36; $p < .01$); by Newman-Keuls analysis, the rats drank more unflavored water TS (M= 5.9 ml) than the novel flavored TS (M= 4.0 ml) and more novel flavored TS than conditioned aversive TS (M= 1.3 ml) with all p's $< .01$. Neither the infusion condition ($F_{(1,66)}$= .20) nor the interaction ($F_{(1,66)}$= 1.16) were significant.

The CS+ conditions were then compared as transformed square root latency scores by a one-way ANOVA for each type of TS. As would be expected by the results of the traditional conditioned suppression experiments, each test solution supported a CR of a different duration ($F_{(2,33)}$= 21.78; $p < .001$); by Newman-Keuls analysis, unflavored water supported a shorter CR than the novel TS ($p < .05$) and the novel TS supported a shorter CR than the conditioned aversive TS ($p < .01$). Thus, as in traditional conditioned suppression of licking, the strength of the CS+ elicited suppressive CR is influenced by the rat's tendency to drink the test solution.

However, note that in Figure 9, the tendency to resume drinking following an exposure to the CSc flavor also differed by the test solution which

was consumed ($F_{(2,33)} = 9.96$; $p < .01$). By Newman-Keuls analysis, the CSc exposure caused greater suppression when the TS was a conditioned aversive solution than when the TS was a novel solution or unflavored water (p's $< .01$); but, there was no difference between the CSc exposed rats which consumed water or novel TS.

Since the latency to begin licking following both the CS+ and the CSc infusion conditions was influenced by the type of TS consumed, the overall square root latency scores of Groups CS+ and CSc on Test Day 1 were analyzed in a 2 x 3 ANOVA. In order to demonstrate that the strength of the suppressive CR was, in fact, determined by the strength of the baseline response tendency, the analysis must reveal a significant infusion condition x TS type interaction; that is, the difference in the degree of suppression seen between Group CS+ and Group CSc should be largest when the TS is aversive (Experiment 6) and should be smallest when the TS is familiar, unflavored water (Experiment 4). As was expected, both the infusion condition ($F_{(1,66)} = 78.42$; $p < .001$) and the TS type ($F_{(2,66)} = 30.50$; $p < .001$) effects were significant: Overall, the CS+ exposed rats were more hesitant to resume

drinking than the CSc exposed rats, and the overall degree of hesitancy was greater for a conditioned aversive TS than a novel TS (N-K, $p<.01$) and greater for the novel TS than a water TS (N-K, $p<.05$). However, the interaction was not significant ($F_{(2,66)}=1.13$). Therefore, the difference between CS+ and CSc elicited suppression was not significantly effected by the type of TS which the rats drank. In the next section, I will explain why this may not differ from the shock situation.

General Discussion of Experiments 4, 5 and 6

Experiments 4, 5 and 6 demonstrated that the suppressive CR elicited by a lithium-paired flavor CS+ is a general phenomenon; it was evident whether the rats consumed water, a novel solution or a conditioned aversive solution. However, the CR is of a relatively short duration because it was most clearly revealed by the rat's hesitancy to return to drink. In fact, when the TS was unflavored water, the rats drank normally once they completed the second lick. When measured as the latency to begin drinking, the duration of the CR in Experiments 4 - 6 ranged from 45 seconds with the unflavored water TS to 235 seconds with the conditioned aversive TS.

Since the duration of the suppressive CR was greatest when the rats consumed the least preferred test solution, the conditioned suppression response elicited by a lithium-paired flavor CS+ is apparently influenced by similar motivational factors as the conditioned suppression response elicited by a shock-paired external CS+. However, the present experiments suggest that the rat's motivation to drink the TS not only effected the

suppression following presentation of the CS+, but also effected the suppression following the presentation of an equally familiar flavor which had not been paired with illness (CSc). When the degree of suppression elicited by the CS+ was compared with the degree of suppression elicited by the CSc, the type of TS did not influence the strength of suppression. It is likely that a similar effect would be evident in the traditional conditioned suppression paradigm. In fact, Ayres and his associates (Ayres, 1968; Ayres and Quincy, 1970) found no difference in the strength of suppressed licking of 8% (low reward) or 32% (high reward) sucrose solution when the number of licks during the shock-paired CS+ exposure was compared with the pre-CS baseline. The preference for the test solution which rats consume determines the tendency to drink, but the overall tendency does not appear to influence the strength of suppression elicited by the CS+ relative to that elicited by either another safe stimulus (CSc) or to the pre-CS baseline.

Finally, the avoidance tests with the discriminative CS flavored solutions indicated that the previous exposures to the CS+ flavor did not weaken

the rats aversions to that flavor, even though, in each experiment, the suppressive CR had weakened considerably by the final suppression test. Thus, the CS+ elicited CR must not be the primary motivator of a rat's avoidance of a flavored solution previously paired with lithium-induced illness. A similar phenomenon has been demonstrated by Kamin, Brimer and Black (1963) using traditional paradigms. They devised a technique which would monitor fear of a shock-paired CS used during avoidance conditioning. The rat was first trained to bar press in an operant conditioning box and then was given avoidance training in a shuttlebox. After a number of successful avoidances to the shock-paired CS+, the rat was placed back in the operant box and the shuttlebox CS+ was presented while the rat responded; the suppression of responding to the CS+ measured the strength of the fear. Kamin et. al. determined that as the number of avoidance acquisition training trials increased, with the rat successfully avoiding the shock with each CS+ presentation, the suppressive CR in the operant box elicited by the CS+ decreased. Thus, as in Experiments 4 - 6 of the current investigation, although the suppressive CR elicited by the CS+

had decreased, the avoidance response to the CS+ had not weakened. Hence, in two very different situations, the avoidance response appears to be motivated by some other factor than simply the CR elicited by the CS+.

CHAPTER V: INFLUENCE OF US AND CS INTENSITY ON THE STRENGTH OF THE SUPPRESSIVE CR

Experiment 7

The strength of traditional conditioned suppression is not only influenced by the strength of motivational factors during testing, but is also influenced by the strength of conditioning factors during training. More specifically, two factors which are positively related to the strength of suppression are the US intensity (e.g., Annau and Kamin, 1961) and the CS intensity (e.g., Kamin, 1965). These factors have also been shown to effect the strength of a conditioned flavor aversion (see Revusky and Garcia, 1970). Experiment 7 was designed to measure the effects of parametric variations in US and CS intensity on the strength of the suppressive CR elicited by a lithium-paired flavor CS+.

The following experiment was partially modelled on an experiment by Nachman and Ashe (1973) which determined the effectiveness of various dosages of LiCl in establishing a flavor aversion to a 15% (w/v) Sucrose solution. By using similar

training procedures and doses of LiCl, the strength of the suppressive CR obtained in Experiment 7 could be roughly compared with the strength of a flavor aversion in the Nachman and Ashe experiment. The doses of .15 M LiCl used in Experiment 7 were 0.3 mEq/kg, 1.2 mEq/kg and 3.0 mEq/kg which ranged from nearly the lowest dose to the highest dose used by Nachman and Ashe. These doses were considerably weaker than the highest dose on the second conditioning day of the previous experiments which was approximately 6.0 mEq/kg of .47 M LiCl.

The CS intensity was also manipulated in Experiment 7; half of the rats consumed a 20% (w/v) Sucrose solution (Strong CS) and half consumed a 10% (w/v) Sucrose solution (Weak CS). During the subsequent suppression test, the rats were infused with the same concentration of sucrose which they consumed during conditioning. Note that this procedure differs from that of Experiment 3. In Experiment 3, the CS intensity differed between the training and the testing phase; all rats were conditioned with the medium strength solution, but were tested with either a high or a low strength solution. Under these conditions, the CS intensity did not influence

the hesitancy to resume drinking by either the experimental or the control rats. Since the CS intensity, per se, does not effect the rats' hesitancy to drink, the control groups in Experiment 7 were not expected to be influenced by variations in the concentration of the CS solution. On the other hand, when the experimental rats were administered a dose of lithium that was intense enough to support conditioning, those rats which consumed the strong CS were expected to show a greater suppressive CR than those which consumed the weak CS.

The various groups in Experiment 7 are presented in Table 13. There were six Sucrose→ LiCl experimental groups: At each CS intensity level (20% or 10% Sucrose) there were three US intensity levels (0.3 mEq/kg, 1.2 mEq/kg and 3.0 mEq/kg). There were also four control groups. The two Sucrose→Saline control groups determined the effect of experience with Sucrose CS alone on the strength of suppression. The two LiCl→ Sucrose Pseudoconditioning control groups controlled for sensitization effects by administration of the 3.0 mEq/kg of LiCl US 5 hours prior to the Sucrose CS; it is clear that such a procedure will not

TABLE 13. The conditioning procedures of Experiment 7 (6 subjects per group).

Experimental Groups

20% Suc → 3.0 mEq/kg LiCl	10% Suc → 3.0 mEq/kg LiCl
20% Suc → 1.2 mEq/kg LiCl	10% Suc → 1.2 mEq/kg LiCl
20% Suc → 0.3 mEq/kg LiCl	10% Suc → 0.3 mEq/kg LiCl

Saline Control Groups

20% Suc → Normal Saline	10% Suc → Normal Saline

Pseudoconditioning Control (Five hr US - CS interval)

3.0 mEq/kg LiCl → 20% Suc	3.0 mEq/kg LiCl → 10% Suc

produce a sucrose aversion (Boland, 1973).

The strength of conditioning was determined by the suppression test procedure. While consuming unflavored water TS, the rats were exposed to the concentration of sucrose with which they had been conditioned. The strength of the suppressive CR was measured by the latency to complete 10 licks of unflavored water immediately after the CS exposure.

Method

Subjects. Sixty male Sprague-Dawley rats weighing between 184 - 225 gms were maintained on a 24 hr water deprivation schedule with water presented for 10 minutes per day throughout most phases of the experiment. Food was available ad-lib, except during baseline training and suppression testing procedures. As in the previous experiments, the rats were surgically implanted with intraoral cannulae and allowed free access to water for the following three days. On the third day, their cannulae were flushed with water.

Pretraining (Days 1 - 3). On each of three days, all rats were given 10 minutes access to water in a bottle and the amount consumed was measured.

Conditioning Trial (Day 4). On Day 4, the rats were conditioned as previously described in Table 13; there were six rats in each group. The rats in the six experimental groups and the two saline control groups were presented bottles containing either 20% (w/v) Sucrose solution (Strong CS) or 10% (w/v) Sucrose solution (Weak CS) for a 10 minute drinking period. Immediately after the bottles were removed, the experimental rats were injected with 4.0 ml (3.0 mEq/kg), 1.6 ml (1.2 mEq/kg) or 0.4 ml (0.3 mEq/kg) of .15 M LiCl and the saline control rats were injected with 4.0 ml of normal saline. An additional twelve pseudoconditioning control rats were given a backward pairing of lithium and sucrose; they were injected with 4.0 ml (3.0 mEq/kg) of .15 M LiCl, five hours prior to a 10-minute presentation of either 10% or 20% Sucrose solution.

Baseline Training (Days 5 and 6). On Days 5 and 6, all rats were given baseline training trials. As in the previous experiments, the food was removed from all cages thirty minutes before

the test. The rats were then presented a bottle of water and, thirty seconds after they began to drink, were given a 2 ml, 15 second infusion of unflavored water. Immediately after the infusion, the latency to complete 10 licks of unflavored water was measured. The bottle was removed ten minutes after it had been presented and the food was returned to the home cage thirty minutes later. There were only two baseline training days because in Nachman and Ashe's (1973) procedure, the model of the current experiment, only two days intervened between the conditioning day and the testing day.

Suppression Test (Day 7). On Day 7, the rats were given the suppression test in a manner similar to the baseline training procedure in unspecified details. Each rat was presented a bottle of unflavored water test solution and, thirty seconds after it began to drink, was infused for 15 seconds with the concentration of sucrose solution with which it had been conditioned. The subsequent latency to complete 10 licks of the water TS was measured. On Days 8 and 9, the rats received 10 minutes of water per day and the amount consumed was measured.

Aversion Test (Day 10). On Day 10, each subject was presented a bottle containing its CS

flavored solution. The amount consumed in ten minutes measured the strength of the aversion in each group.

Design and Data Analysis. Although the overall design of Experiment 7 was a 2 x 5 (CS Condition X US Condition), the control groups and the experimental groups were analyzed separately; the control groups were analyzed as a 2 x 2 ANOVA and the experimental groups were analyzed as a 2 x 3 ANOVA. The controls were then compared with each of the experimental groups by individual t-tests.

The results of both the suppression test and the aversion test were transformed into suppression ratios to reduce individual variability within the groups. A latency suppression ratio was computed as described in Experiment 2 ($\frac{\text{Test Latency}}{\text{Test Latency + Baseline Lat.}}$). In order to obtain a suppression ratio for the results of the aversion test, the sucrose intakes were first transformed into preference ratios. The amount of sucrose consumed on the aversion test day was divided by the sum of the sucrose intake on the test day and the water intake on the preceding baseline training day ($\frac{\text{Sucrose}}{\text{Sucrose + Water}}$). These preference ratios were then subtracted from 1.0 to obtain suppression ratios

which could be graphically compared with the results of the suppression test. Thus, a value of 1.0 means complete suppression and a value of 0.5 means equal preference of water and sucrose.

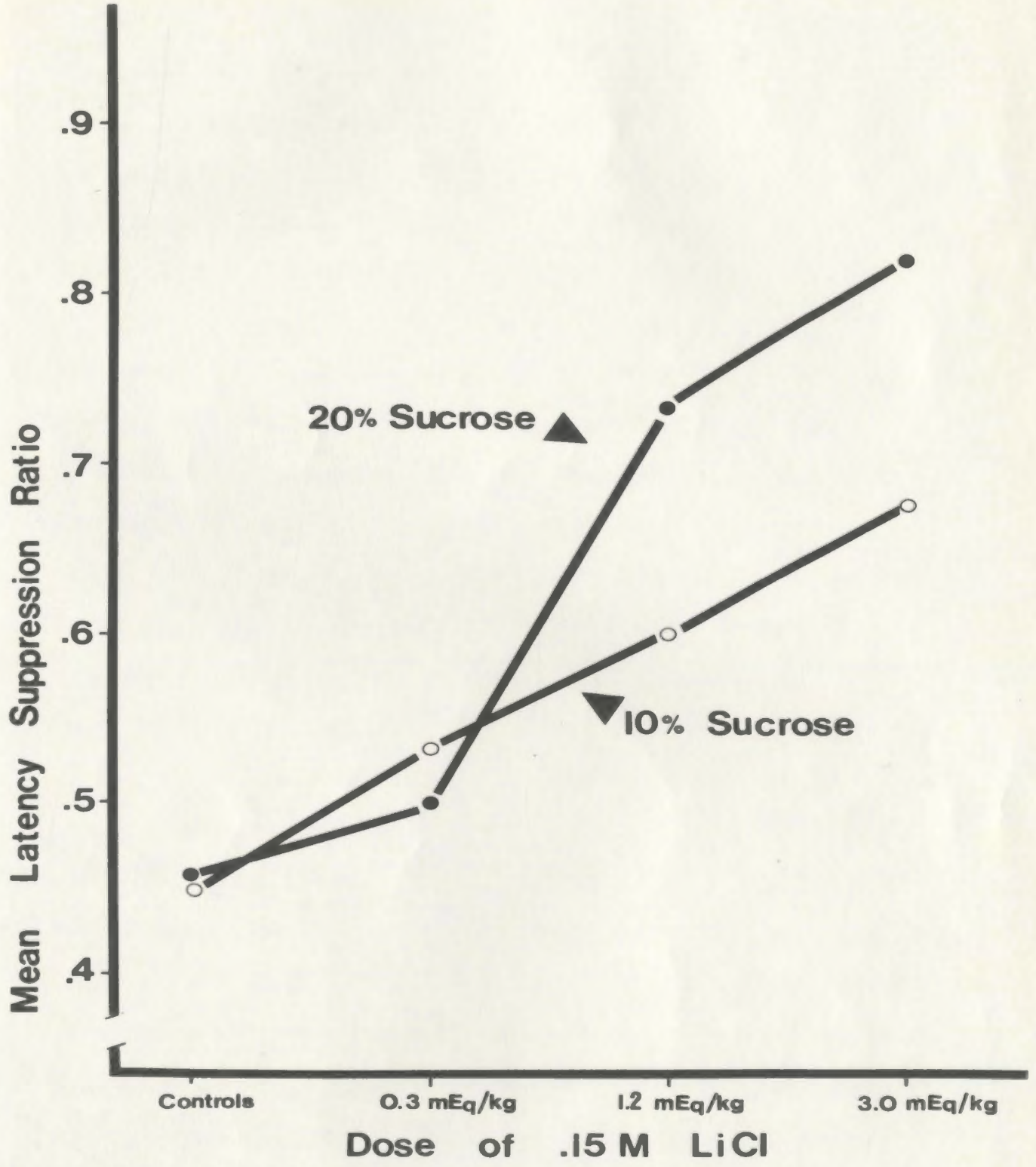
Results

Suppression Test. Figure 14 presents the mean latency SR for each group in Experiment 7. The data points to the far left represent the pooled controls at each CS intensity level. A 2 x 2 ANOVA for each control condition at each CS intensity revealed no differences (Control condition: $F_{(1,20)}$= 1.63; CS intensity: $F_{(1,20)}$= .01; Interaction $F_{(1,20)}$= 1.24).

The remaining data points in Figure 14 represent the experimental groups. As is suggested by the figure, the US intensity influenced the strength of the suppressive CR ($F_{(2,30)}$= 3.62; $p<.05$); a trend analysis indicated that the CR increased linearly with the dose of lithium administered during conditioning ($F_{(1,30)}$= 7.06; $p<.01$). However, neither the CS intensity ($F_{(1,30)}$= 1.38) nor the CS x US interaction ($F_{(2,30)}$= .74) were significant.

Each of the experimental groups was then compared with the pooled controls at each level

Figure 14. Mean latency suppression ratio for the groups in the suppression test of Experiment 7.


Mean Latency Suppression Ratio
.9
.8
.7
.6
.5
.4
20% Sucrose
10% Sucrose
Controls
0.3 mEq/kg
1.2 mEq/kg
3.0 mEq/kg
Dose of .15 M LiCl

of sucrose concentration. There were thus 12 comparisons: The two control conditions were compared with each of the six experimental conditions. Neither of the groups which were conditioned with the lowest US dose (0.3 mEq/kg) differed from either control condition (t's (16) $<$ 1.0). Of the two groups which were conditioned with the middle US dose (1.2 mEq/kg), only the group which consumed 20% sucrose showed a suppressive CR when compared with both control groups (t's (16) $>$ 2.51; p's $<$ .025); however, the rats which consumed 10% sucrose did not differ from controls (t's (16) $<$ 1.48; p's $>$ .05). Finally, at the highest US dose (3.0 mEq/kg), both the 20% and the 10% sucrose supported conditioning (t's (16) $>$ 2.27; p's $<$ .025).

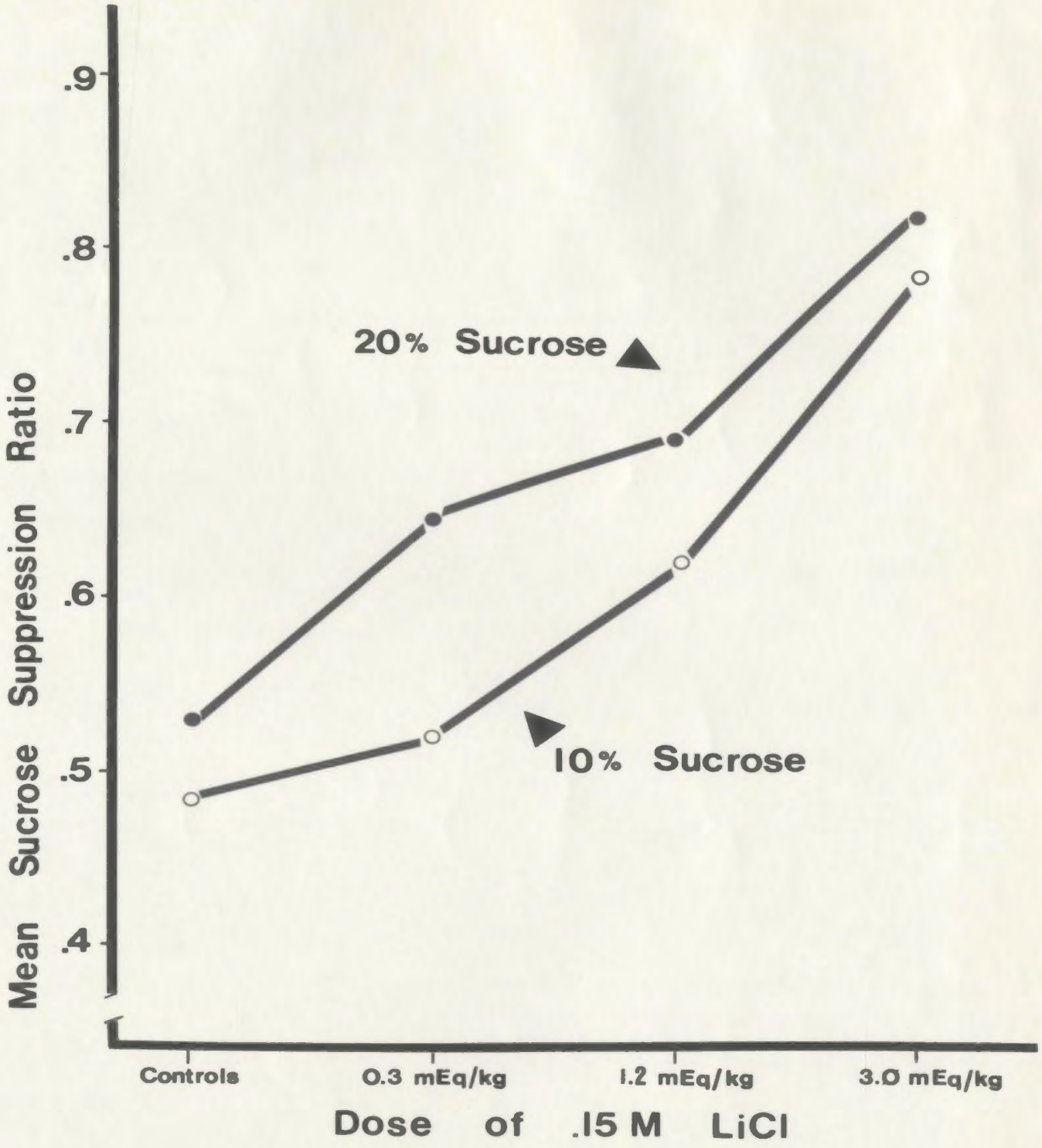
Although the CS x US interaction in the 2 x 3 ANOVA of the experimental groups was not significant, the CS intensity groups were compared at the pooled US intensity levels which supported conditioning. There were two reasons for this additional analysis: 1) It had been predicted on an <u>apriori</u> basis that the strength of the suppressive CR would vary by the strength of the CS+ flavor consumed at only those US intensity levels which supported conditioning. 2) At the 1.2 mEq/kg dose level, the 20% sucrose

group showed evidence of suppression when compared with the controls, but the 10% sucrose group showed no such CR. It appeared that at the medium dose level, the intensity of the CS flavor determined whether or not the rats would develop the association. The comparison between the two CS intensity groups pooled across the 1.2 mEq/kg and the 3.0 mEq/kg dosage levels indicated that the strong CS elicited a greater CR than the weak CS ($t(22) = 1.73$; $p < .05$, one-tailed). Thus, the CS intensity did have an influence, although it was statistically marginal.

Aversion Test. Figure 15 presents the mean sucrose suppression ratio for each group in Experiment 7; the greater the suppression ratio, the stronger the aversion. The first two data points represent the pooled control conditions at each CS intensity level. A 2 x 2 ANOVA for control condition and CS intensity level indicated that there was no difference in the preference for 10% or 20% sucrose by the control groups (Control Condition: $F_{(1,20)} = 0.61$; CS Intensity: $F_{(1,20)} = 2.74$; $p > .05$; Interaction: $F_{(1,20)} = 1.41$).

The remaining data points represent the experimental groups. A 2 x 3 ANOVA showed a significant US intensity effect ($F_{(2,30)} = 18.32$; $p < .01$) and

Figure 15. Mean Sucrose suppression ratio of each group in the aversion test of Experiment 7. The scores were first transformed into preference ratios relative to the amount of water consumed on the previous baseline day ($\frac{\text{Sucrose}}{\text{Sucrose + Water}}$). These preference ratios were then subtracted from 1.0 to obtain suppression ratios that could be graphically compared with the latency suppression ratios of the previous suppression test.


Mean Sucrose Suppression Ratio
.9
.8
.7
.6
.5
.4
20% Sucrose
10% Sucrose
Controls
0.3 mEq/kg
1.2 mEq/kg
3.0 mEq/kg
Dose of .15 M LiCl

a trend analysis indicated that the increase in CR strength was linear across dosages ($F_{(1,30)}$ = 30.43; $p < .001$). In addition, the rats showed a greater aversion to the 20% sucrose (Strong CS) than to the 10% sucrose (Weak CS) with $F_{(1,30)}$ = 5.26 ($p < .01$); however, the CS x US interaction was not significant ($F_{(2,30)}$ = 0.26).

The sucrose suppression ratios of the pooled controls were then compared with those of the experimental groups. Each experimental group showed a greater aversion to sucrose than the control groups at either CS intensity level (t's (16) $>$ 1.85; p's $<$ .05, one-tailed), except the Weak CS - 0.3 mEq/kg US group which did not differ from either control group (t's (16) $<$ 1.50; p's $>$ .05).

Discussion

As in the traditional conditioned suppression paradigm, the strength of the suppressive CR elicited by a lithium-paired flavor CS+ was influenced by the US intensity during conditioning. Although the CS intensity effect was weak, the results suggest that this parameter also influences the strength of the CR. This effect, presumably, is the result of a stronger association established during

conditioning, since CS intensity changes did not influence the performance of the CR in Experiment 3.

The aversion test, which was administered three days after the suppression test, appeared to be a more sensitive indicator of a flavor-illness association than the suppression test. This was suggested by the finding that both the Strong CS-low dose (0.3 mEq/kg) and the Weak CS - medium dose (1.2 mEq/kg) experimental groups demonstrated a significant aversion to sucrose, but showed no significant suppressive CR. Incidentally, Nachman and Ashe (1973) reported that the 0.3 mEq/kg dose was effective in establishing an aversion to 15% Sucrose solution, which supports the present results.

CHAPTER VI:
GENERAL DISCUSSION

A basic premise of the preceding experiments was that a flavor which is paired with a toxic agent, lithium chloride, gains the capacity to elicit a conditioned sickness response in rats. Many species show the clearcut sickness CR of vomiting when presented a conditioned aversive substance (see Garcia, Rusiniak and Brett, 1977), but rats are incapable of vomiting and thus do not show such obvious behavioral evidence of conditioned sickness. Therefore, it was necessary to devise an indirect test of sickness to measure the CR elicited by an exposure to a lithium-paired flavor. The Sickness UR,itself, suppresses consumption of novel flavored solutions (Domjan, 1977; Green, McGowan, Garcia and Ervin, 1968) and of unflavored water (Haroutunian, Riccio and Gans, 1976; also see Appendix A). Under the working assumption that the sickness CR would resemble the sickness UR, I measured the capacity of the lithium paired flavor CS+ to suppress consumption of a test solution different in flavor from CS+.

Similarity to the traditional paradigm with shock

The measure of the CR in the present series of experiments is similar to the measure employed in the traditional paradigm with shock; that is, suppression of ongoing appetitive responding. According to Pavlovian stimulus substitution theory, a flavor CS+ ought to elicit a conditioned sickness response as a result of having been paired with sickness, just as an external CS+ ought to elicit a conditioned fear response as a result of having been paired with shock. Both of these CRs ought to interfere with appetitive responding, and, indeed, I found little difference between the ostensible conditioned sickness response and the usual conditioned fear response. Not only do both sickness and fear CRs produce the same suppressive effect on ongoing appetitive responding, but they also appear to follow similar laws of conditioning, since variations in CS+ and US intensity during conditioning effect both CRs in a similar manner.

Motivational factors at the time of testing also influence the strength of the suppressive CR in both paradigms; the more highly preferred the type of test solution we use, the weaker the CS+ elicited suppressive CR (e.g., Vogel and Spear, 1966). However,

the motivational factors during the suppression test not only influenced the strength of suppression elicited by the CS+, but also influenced the strength of suppression elicited by the CSc. In fact, when the suppressive CR was defined as the difference in the duration of suppression between the CS+ and CSc conditions, the preference for the test solution did not have a statistically significant effect on the strength of suppression. It seems likely that a similar conclusion would be drawn about the traditional conditioned suppression CR with a shock US, but, unfortunately, the exact parallel experiment with shock has not yet been done.

Is the suppression caused by a sickness CR?

The term "sickness CR" suggests that the CS+ paired with sickness produces a CR which is similar to sickness; however, this suggestion is not supported by my results. Although exposure to the lithium-paired flavor CS+ repeatedly suppressed consumption of the test solution, the rats response patterns during the suppression interval did not resemble the response patterns of sick rats. When rats are injected with lithium chloride, they become lethargic, show depressed responding and lie on the floor of their

cages. On the other hand, when rats were infused with the lithium-paired flavor CS+, they often demonstrated the following behavioral sequence: 1) Freezing during the infusion and shortly thereafter, 2) Agitation, including facewashing, as if to remove the aversive substance from their mouths, and moving in a short-jerky manner in their cages, 3) Freezing toward the back of their cages, and finally 4) Cautiously approaching the bottle containing the test solution to resume drinking.

Not only are the behavioral indicators of the CR different from those of the lithium-induced UR, but also the duration of the suppression is shorter for the CR. As measured by the suppressed intake of a novel flavored solution, the sickness UR is maintained for approximately 60 minutes (Domjan, 1977), but the sickness CR is maintained for only approximately three minutes (as shown in Experiment 5). Since the CR elicited by a flavor CS+ differs appreciably from the lithium-induced sickness UR, it may be incorrect to attribute the CS+ elicited suppression of drinking to a conditioned sickness response.

The concern with the underlying cause of suppressed responding elicited by a lithium-paired

CS+ is reminiscent of the concern with the role of conditioned fear in the traditional conditioned suppression paradigm. The presentation of a shock-paired CS+ results in suppression of ongoing responding, presumably, because the CS+ elicits a conditioned fear response. However, the CR to a shock-paired CS+ differs considerably from the UR to shock. The UR elicited by shock to the feet in a confined space is flinching and jumping, but the CR elicited by a shock-paired CS+ in a confined space is freezing and crouching (Blanchard and Blanchard, 1969; Mackintosh, 1974). Hence, a fear CR is not identical to a fear UR, just as a sickness CR is not identical to a sickness UR.

Since in both the traditional and in the present conditioned suppression paradigms, the CR is different than the UR, the terms of "conditioned fear" and "conditioned sickness" are not exact descriptions; they have been used here simply because many others have used them in the past. However, both conditioned suppression effects can reasonably be attributed to classical conditioning regardless of the underlying cause of the CR; in both cases, the response elicited by the CS changed as a result of prior exposure to the CS - US relation and this defines classical

conditioning. It is now clear that the classical CR need not be a replica of the UR; in fact, it is often reported to occur in the opposite direction (e.g., Siegel, 1972).

The role of the CR in the typical flavor aversion test.

The final question to be considered is whether the CR demonstrated here underlies learned taste aversions. To answer this, the similarities and the differences between the conditioned suppression and the taste aversion procedure will first be delineated. On the basis of this analysis, I will describe properties which the CR must have if it is to be deemed responsible for the learned flavor aversion. I will then show that the CR does not have these properties and hence cannot be held responsible for the flavor aversion.

In the conditioned suppression procedure, the CS+ flavored solution is briefly presented, but the test of its effect is on the consumption of a different test solution (TS). In the flavor aversion procedure, the CS+ flavored solution is presented throughout, and the test of its effect is on the consumption of that same CS+ flavored solution. In other words, the difference between the suppression and the aversion

procedure lies in the nature of the TS solution: In the suppression procedure, it is different from CS+, while in the aversion procedure, it is the same as CS+.

For suppression to account for aversions, this difference between suppression and flavor aversion procedures must explain why aversions are longer lasting than suppression; we have seen that suppression lasts under five minutes, while it is known that aversions last many hours. If this difference is to be explained in terms of procedural differences, the explanation would be that when the TS is the same as the CS+ solution, as in the aversion procedure, each lick from the bottle reinstates the CS+ and hence reinstates the suppressive CR. There is no similar reinstatement in the conditioned suppression procedure. If this explanation is accurate, a flavor aversion is merely a product of summed suppressive CRs which occur during the avoidance test period.

In contradiction to this possible explanation of flavor aversions in terms of conditioned suppression is the fact that a dose of lithium too low to produce a suppressive CR produced a conditioned flavor aversion in Experiment 7. Furthermore, in each of Experiments 4, 5 and 6, the suppressive CR was shown to be less

resistant to extinction than was the avoidance response. After three to five suppression extinction trials, the flavor CS+ no longer produced suppression, but when the animals were later given an avoidance test with the same CS+ flavored solution, their aversions had not even weakened. Since the avoidance test appears to be more robust than the suppression test, it is unlikely that the CR elicited by the lithium-paired flavor CS+ is the sole motivator of a flavor aversion. These results seem to justify the preference of Revusky and Garcia (1970) for categorizing learned flavor aversions as due to punishment (passive avoidance) rather than as simply due to classical conditioning.

The finding that the "sickness CR" does not solely account for a conditioned flavor aversion is similar to the finding in the traditional paradigm with shock that the "fear CR" does not solely account for the avoidance response. When rats have acquired a shuttlebox avoidance response, they continuously cross the hurdle with each CS+ presentation; the response does not weaken even though the animals no longer experience shock. When Kamin, Brimer and Black (1963) tested the capacity of the same shuttlebox CS+ to suppress ongoing responding in a Skinner Box,

they found that as the number of successful avoidance trials increased (up to 27 trials), the capacity of the CS+ to suppress responding decreased. It is thus unlikely that the CR elicited by either the lithium-paired flavor CS+ or the shock-paired external CS+ exclusively underlies an avoidance response.

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APPENDIX A

Experiment A. Suppression of unflavored water drinking by a lithium-induced sickness UR.

Experiment A: Suppression of Unflavored Water drinking by a lithium-induced sickness UR

The CR elicited by a lithium-paired flavor CS+ caused rats to suppress their drinking of unflavored water in Experiment 1[a] of the present series of experiments. However, the UR of lithium-induced sickness does not cause suppression of water drinking when measured in each of 10 minute intervals over a period of 60 minutes (Domjan, 1977). Although the temporal parameters grossly differ between the conditioned and the unconditioned responses, according to Pavlovian stimulus substitution theory, both responses are presumably the result of the same underlying process, that is, lithium-induced sickness.

It is conceivable that lithium will suppress drinking of unflavored water when the drinking period is of a shorter duration than that used by Domjan (1977). In fact, Haroutunian, Riccio and Gans (1976) reported that the sickness UR induced by prior rotation suppresses intake of unflavored water when measured as the latency to begin licking. Therefore, the following experiment measured the effect of lithium-induced illness on the amount of unflavored water consumed in a five and a ten minute drinking period. The 3.0 mEq/kg dose of .12 M LiCl was

equal to the highest dose used by Domjan (1977).

Method

Subjects. The subjects were 16 male Sprague-Dawley rats ranging between 358 - 435 gms on ad-lib access to Purina rat chow except during the 10-minute drinking period each day.

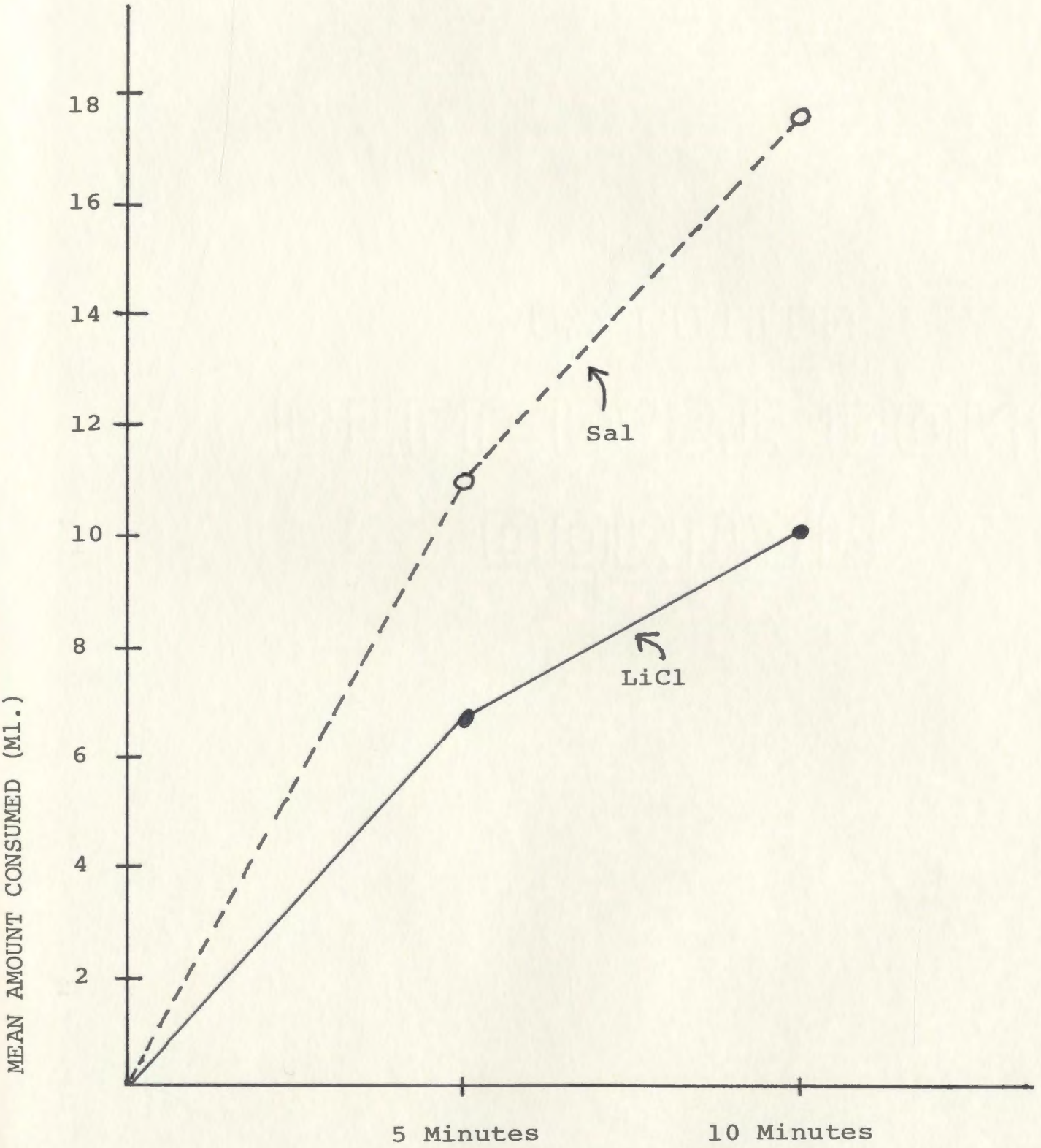
Procedure. The rats were initially adapted to drinking water for 10 minutes per day for six days. On the seventh day, they were tested.

On the test day, half of the rats were injected with 3.0 mEq/kg of .12M LiCl and the other half were injected with an equal volume of saline. Thirty minutes after they were injected, the rats were presented unflavored water in a bottle. The amount consumed after 5 minutes and after 10 minutes was measured.

Results and Discussion

The lithium UR resulted in suppressed water drinking during both the first five minutes ($t(22)=5.36$; $p<.001$) and during the complete 10 minutes ($t(22)=6.76$; $p<.001$) of drinking as shown in Figure 1. These results suggest that the UR of lithium-induced sickness not only suppresses the consumption of novel flavored solutions, but also of unflavored water when measured by a brief drinking test.

Figure 1. Mean amount consumed of unflavored water by rats previously injected with LiCl (Li) or Saline (Sal).


MEAN AMOUNT CONSUMED (Ml.)
18
16
14
12
10
8
6
4
2
Sal
LiCl
5 Minutes
10 Minutes

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APPENDIX B

Generalized Conditioned Flavor Aversions

Generalized Conditioned Flavor Aversions

Wild rats (Barnett, 1963; Richter, 1953; Rzoska, 1954) as well as laboratory rats (e.g., Best and Batson, 1977; Carrol, Dinc, Levy and Smith, 1976; Domjan, 1976; Green and Parker, 1975; Nachman and Jones, 1974; Revusky and Bedarf, 1967; Siegel, 1974) exhibit a tendency to avoid novel foods. This tendency, called neophobia, has been shown to be enhanced when rats have previously experienced illness following food consumption (Carrol, Dinc, Levy and Smith, 1976; Domjan, 1975; Richter, 1953; Rozin, 1968; Revusky, Parker, Coombes and Coombes, 1976; Rzoska, 1954), but not when rats have previously experienced illness in the absence of prior food consumption (Best and Batson, 1977; Domjan, 1975; Revusky, Parker, Coombes and Coombes, 1976). Domjan (1975) has suggested that in the former cases, an aversion to the flavor which preceded illness generalizes with the novel flavor, whereas in the latter cases, there is no opportunity for such stimulus generalization. When rats were injected with a toxin following consumption of saccharin, they drank less of a novel casein

solution than did rats injected with a toxin in the absence of prior consumption (toxicosis alone), this indicated stimulus generalization between saccharin and casein. There was no similar stimulus generalization between saccharin and vinegar.

Enhanced neophobia following a food-illness pairing was systematically investigated by Rozin (1968). On the conditioning day, the experimental rats became ill after eating a greasy-bland diet; the controls consumed the same diet but did not become ill. In a subsequent choice test between a safe familiar diet or a novel diet, all rats demonstrated neophobia; however, the neophobia was more pronounced in the experimental rats than in the control rats. On the other hand, Brackbill, Rosenbush and Brookshire (1971) failed to demonstrate enhanced neophobia in another similar paradigm. This difference may be explicable in terms of stimulus generalization in Rozin's results and its absence in those of Brackbill, et al. Historically, stimulus generalization has been explained in terms of common elements between stimuli. Rozin used diets which were bland-greasy (CS+), bland-fine

powdered and sweet-granular; conceivably, an aversion to the bland-greasy diet may have generalized to the bland-fine powdered diet, as suggested by Rozin's report that all rats demonstrated a greater neophobia to the bland-powdered diet than to the sweet-granular diet (although he reported no diet by treatment interaction). However, Brackbill, Rosenbush & Brookshire used three flavored solutions selected from the differing taste categories of sweet (saccharin), salty (NaCl) and sour (Citric Acid); presumably, since these independent flavors had few common stimulus elements, there was little opportunity for generalization of an aversion from the toxicosis-paired flavor to the novel flavor.

Stimulus generalization between flavors, then, may result from overlap of common elements of a toxicosis-paired flavor and a novel flavor and hence result in apparent enhanced neophobia after a poisoning experience. Psychophysical experiments have attempted to separate gustatory stimuli into four independent categories--sweet, sour, salty and bitter; a "pure" flavor,

containing elements exclusively from a single category, would not be expected to generalize with a flavor lacking elements of that category. However, the flavors employed in flavor aversion learning experiments are typically not "pure"; for instance, saccharin (in high concentrations), a common CS, has both sweet and bitter components to human judges. The present study attempted to delineate a set of flavors which show minimal generalization with one another from a larger set of flavors commonly employed in experiments of flavor aversion learning. Such "orthogonal" flavors, might be useful as stimuli in experiments which employ multiple CSs.

Since the designs of the following two experiments were complex, we will outline the logic of Experiment 1 here. Rats were divided into 10 training groups of nine rats each; these groups were trained through contingent lithium injections to have extremely strong aversions to solutions of (1) casein, (2) coffee, (3) grape juice, (4) milk, (5) quinine, (6) saccharin, (7) sucrose, (8) vinegar, (9) NaCl, or (10) were subjected to toxicosis alone (that is, injected with lithium in the absence of prior consumption

of a flavored solution). Each rat within these groups was subjected to a different testing sequence in which it consumed each of these nine flavored solutions once in a one-bottle test. The net result was that one rat from each training group drank casein on the first day, another rat drank it on the second day, and so on in a balanced design. More specifically, the basic sequence was casein, coffee, sucrose, vinegar, NaCl, milk, grape juice, quinine, saccharin. Variants of this sequence were derived by beginning with a different flavor than casein and continuing to the end of the basic sequence described above. For instance, if a rat began with a milk test, its sequence was milk, grape juice, quinine, saccharin, casein, coffee, sucrose, vinegar, NaCl. Each rat of the nine rats in a training group began testing with a different one of the nine flavors. This procedure produced two measures of generalization between Flavors A and B. One was based on the preference for Flavor B among rats trained to have an aversion to Flavor A and the other was based on the preference for Flavor A among rats trained to have an aversion to Flavor B.

EXPERIMENTS 1 AND 2

Method For Experiment 1

Subjects. Ninety male Sprague-Dawley rats, weighing 218-248 gms, ad-lib, were individually housed in stainless steel cages and only removed when weighed or injected.

Flavored solutions. Nine solutions were used: 5% (w/v) enzymatic Casein Hydrolysate, 1.25% (w/v) Sanka de-caffeinated Coffee, 50% (v/v) evaporated Milk, 50% (v/v) Welch's Grape Juice, .01% (w/v) Quinine Sulfate, 0.4% (w/v) Sodium Saccharin, 20% (w/v) Sucrose, 3% Cider Vinegar and 1.5% (w/v) Sodium Chloride (NaCl).

Conditioning Trials. (Days 1,2,& 5). Following three days of 10 minutes access to water in a bottle, the rats were conditioned. Nine rats were assigned, matched by body weight, to each of the ten groups previously described.

There were three conditioning trials (Days 1, 2 and 5). On each trial, an experimental rat was presented a bottle containing the appropriate flavored solution for 10 minutes. As soon as the bottle was removed, the rat was injected intraperitoneally with 2% w/v (.47M) Lithium

Chloride (LiCl) in solution with distilled water; the doses on Days 1,2 and 5 were 1, 2 and 3 ml respectively. Nine control rats (toxicosis alone) were injected with the same volume of LiCl as the experimental rats, but did not drink earlier. Instead, they were presented water for ten minute period which began four hours after the LiCl injection; under such conditions, a learned aversion to unflavoured water ought not to develop. (eg. Boland, 1973; Barker and Smith, 1973). On Conditioning Days 2 and 5, all rats were given 18 hours of water from a bottle 1.5 hours after the injection of LiCl; there was no additional water presented on Conditioning Day 1. On a given trial, if a rat drank less than 3 ml, it received a 2 ml infusion of the conditioning solution, washed across its tongue through a syringe in the course of about 10 seconds, prior to the LiCl injection. After the final 18 hr. access to water, the rats were maintained on 12 minutes water per day for three days (Days 7, 8 and 9).

Testing Trials (Days 10-27). Nine test trials occurred on alternate days over an 18 day period. On each trial, the rats received 12

minutes per day access to the appropriate test solution; on the intervening day, they received 12 minutes access to water. The sequence of test solution presentation was as previously described in the introduction.

On Test Day 1, each rat from a given training (CS+) group drank a different test solution (TS); consequently, each test solution was consumed by ten rats, one from each of the nine CS+ groups and one control. The same procedure was followed on each of the remaining eight test days, according to the TS presentation sequence. By the completion of the nine days, all rats had been exposed to each of the test solutions (including its own CS+ solution); however, one subject from each group began at a different point in the TS presentation sequence. Thus, for instance, there were to be nine tests of casein on rats made sick after drinking sucrose; one each of these tests was to be on each of the days of testing. The TS preference scores of a given CS+ group were then pooled across test days. Of course, the rats would be expected to show stronger aversions in the earlier tests than in the later tests, because each test trial serves as an extinction trial

when the CS+ flavor and the TS flavor share common elements. However, this effect of generalized extinction was minimized by establishing extremely strong aversions during conditioning and any extinction effects were counterbalanced among training groups. In addition, we controlled for this extinction in the data analysis, as will be explained later.

An unfortunate experimental error occurred on the first test day: Animals that were scheduled to receive NaCl were actually administered 1.5% Sodium Saccharin. Following the error, the sequence of test solution presentation was altered such that no other rats were tested with NaCl. The data from the group in error were excluded from the data analysis. Thus in Experiment 1 we only have generalization scores between NaCl and other flavors for animals trained on NaCl and tested on the other flavor; we have no data on the NaCl preferences of rats trained with other flavors.

Method for Experiment 2

The procedures of Experiment 2 were identical to those of Experiment 1 except as indicated below. The subjects were 110 male Sprague-Dawley rats

weighing between 215-240 gms, ad-lib.

The groups differed from those of Experiment 1 in two ways: 1) To insure a relatively pure sour stimulus, a 1.5% normal HCl solution was added to the array of flavors. 2) The quinine solution concentration was reduced to .005% to overcome a floor effect that was evident in the quinine intake of control rats in Experiment 1.

Each CS+ group and the toxicosis alone control group contained 10 rats. The conditioning trials and assignment to test conditions proceded exactly as in Experiment 1, except for use of a different sequence of test solution presentation as follows: Casein, milk, grape juice, quinine, HCl , saccharin, NaCl, coffee, sucrose, vinegar. The testing procedure differed slightly. Rather than alternating test days and water days, two water days intervened between each test day to minimize differences in thirst levels between pairs of tests. However, it will become apparent below that the additional day of water did not change the pattern of results that were seen in Experiment 1.

Data Procedure for both Experiments

There were two measures of generalization between two flavored solutions, A and B. A preference for Solution B among rats which had been trained to avoid Solution A and a preference for Solution A among rats which had been trained to avoid Solution B. For instance, in the analysis of casein - sucrose generalization, one measure is the preference for casein of rats made averse to sucrose and a second measure is the preference for sucrose of rats made averse to casein. Both measures were included to illustrate symmetrical generalization effects. However, these raw preferences were not a pure measure of stimulus generalization because there are differences in normal preferences of rats for these different solutions: Rats without any learned aversion will have far higher preferences for sucrose solution than for casein solution. It was necessary to correct for differences in these baseline preferences. Hence, we used the preferences obtained among the rats who did not drink a flavored solution prior to toxicosis (toxicosis alone) during training in order to adjust the

obtained preferences. More specifically, a two step procedure was used to obtain our measures of stimulus generalization as shown in Table 1 and described below.

(1) We first converted all the amounts of flavored solutions consumed during tests into preference measures which adjusted for individual differences in fluid consumption but did not adjust for differences in normal flavor preferences. These preferences (or Kamin-suppression ratios) were $\frac{TS}{TS + W}$, where TS was the amount of the test solution consumed during a one-bottle test, while W was the amount of unflavored water consumed on the preceding day. This is a standard preference measure in flavor aversion literature in which 0.5 indicates that the amount consumed of the test solution was the same as the amount consumed of unflavored water, while 0.0 indicates complete failure to consume the test solution.

(2) We will now define two types of TS/TS+W preference ratios. X ratios are obtained from experimental rats (which had been subjected to a pairing of a flavor with toxicosis). Y ratios are obtained from control rats (which were subjected

Table 1. Method of conversion of raw scores to CPRs.

1) UNCORRECTED PREFERENCE RATIO

TS= Test solution intake
W = Water intake on previous day

Preference ratios:

$X = \frac{TS}{TS + W}$ for the experimental rat

$Y = \frac{TS}{TS + W}$ for the control rat which consumed the same TS as the experimental rat on the same day, but had been subjected to toxicosis alone during training.

2) CORRECTED PREFERENCE RATIO (CPR) $= \frac{X}{X + Y}$

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to unpaired toxicosis during training and hence ought not to exhibit a learned flavor aversion). Y ratios were matched to X ratios on the basis of both the flavor during testing and the day of testing. For instance, suppose a rat which had consumed casein solution prior to toxicosis was tested with saccharin solution on the fifth test and an X ratio was obtained; the corresponding Y ratio would be obtained from a rat which had been subjected to unpaired toxicosis during training and was tested with saccharin solution on the fifth test day. The X ratio was then converted to a corrected preference ratio (CPR) on the basis of the equation CPR = X/X+Y. Note that if the CPR was 0.5, the experimental rat would have exhibited the same preference as the control rat and hence one would suppose that there is no generalization between the training flavor and the test flavor. CPRs reliably below 0.5, indicate generalization between the two test flavors. Such CPRs adjusted for differences in baseline preferences of rats for different flavors so that we could measure generalization effects between any two flavors. If raw scores had been used, our data would not be interpretable.

Note that the Y-score used in the CPR formula was obtained from only a single control rat which had been subjected to exactly the same test sequence as the experimental rat which yielded the X-score. We did not use the mean of the Y scores from control rats subjected to all sequences because we wanted both the X and Y components of the CPR to be based on the same prior history with different flavored test solutions. Among the experimental rats, there was an increase in preference over test trials which resulted from some extinction of the generalized aversions. There was a similar, but less mark increase in preference among the controls due to generalized loss of neophobia. Therefore, using the scores of experimental and control rats with similar test histories in the CPR ratio, $\frac{X}{X + Y}$, reduced changes in the CPR over test trials.

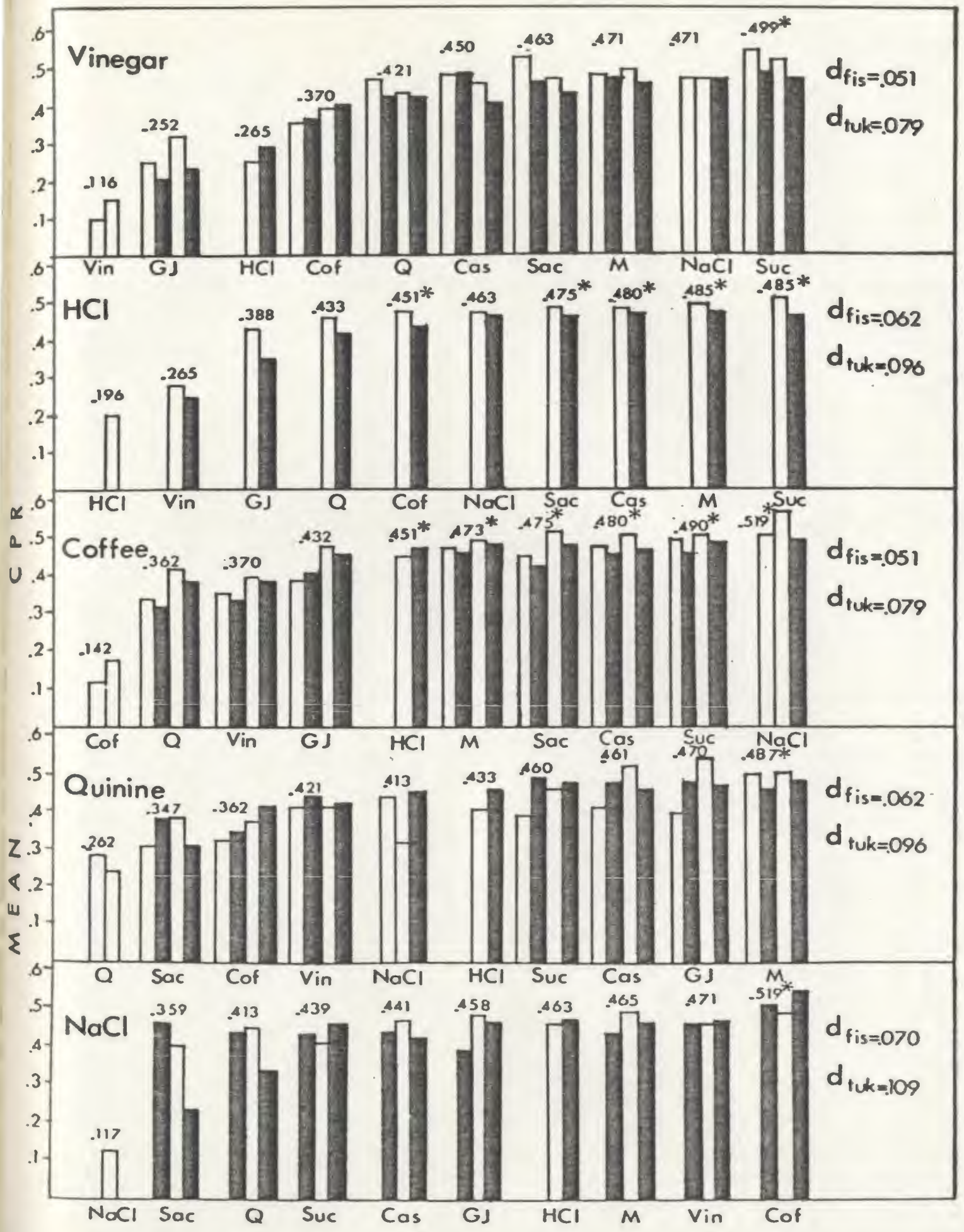
Using only a single Y-score from a single control rat rather than the mean Y-score of all control rats also prevented statistical inter-dependence which would result if all scores were based on a common Y-score. For instance, if a common mean Y-score was used as an input into,

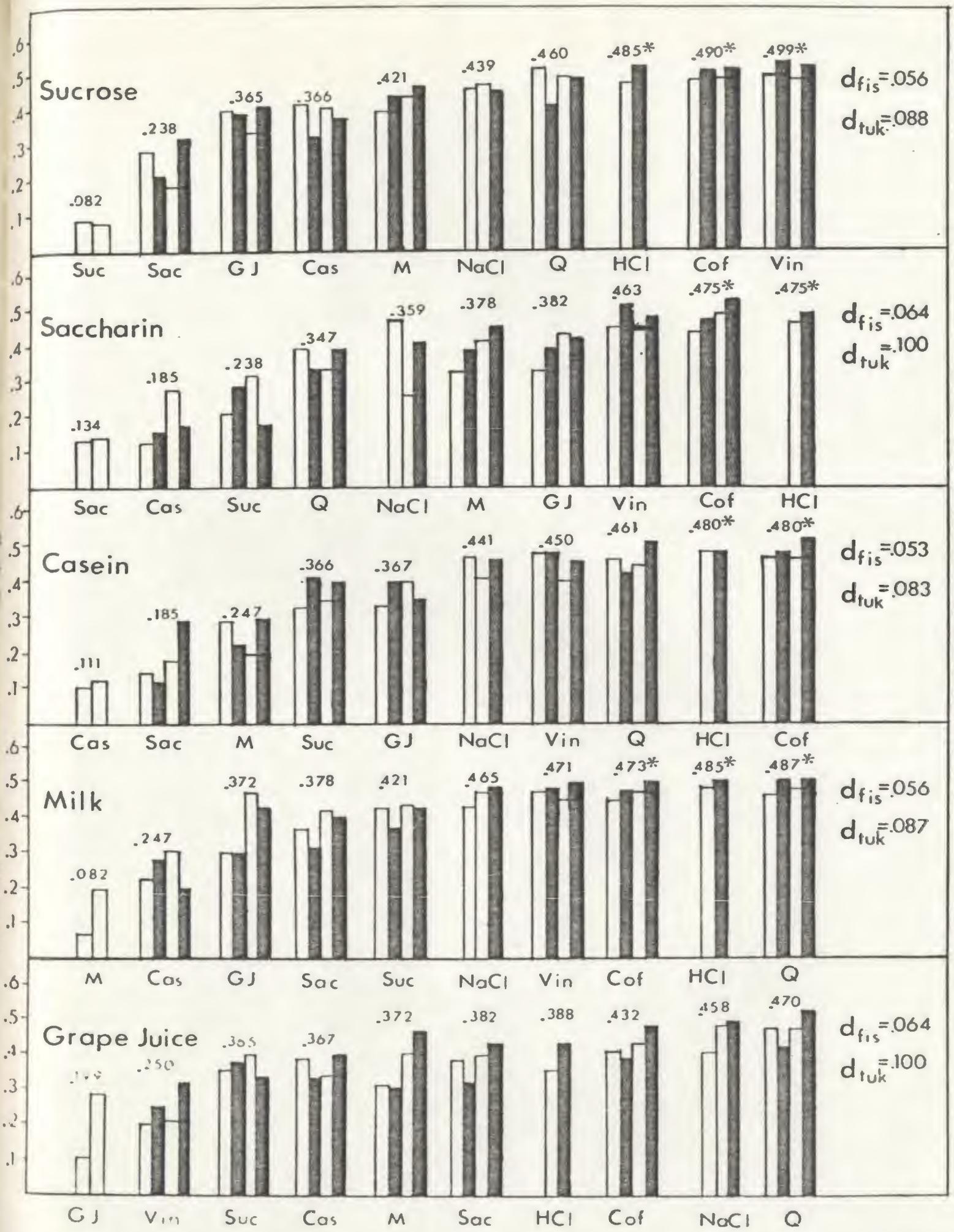
say, 9 vinegar-HCl CPRs and these were compared with 9 vinegar-Coffee CPRs based on a different common mean Y-score, the resulting nonrandomness would invalidate any inferential statistics. The use of single Y-scores excluded such statistical problems. It is true that each Y-score was used to control for a number of X-scores. For instance, the Y-score for saccharin on the fifth test day, controlled for all X-scores for saccharin on the fifth test, regardless of the training flavor. But this does not introduce statistical interdependence when two sets of CPRs are compared.

Results

Figures 1 and 2 show the data for each flavor in terms of CPRs arranged in order of magnitude combined over both experiments. Each row of bars refers to a different flavor. The one or two extreme left bars refer to the case in which the animal is trained and tested on the same flavor. In each case in which there is more than one bar, the left bar refers to Experiment 1 and the right bar refers to Experiment 2. The remaining bars for each flavor differ as to whether they are open or filled. In the case of open bars, the

Figures 1 and 2. Mean TS flavor CPRs between any two flavors in Experiments 1 and 2. See text for explanation.


MEAN CPR
Vinegar
.116 .252 .265 .370 .421 .450 .463 .471 .471 .499*
Vin GJ HCl Cof Q Cas Sac M NaCl Suc
d_{fis}=.051
d_{tuk}=.079
HCl
.196 .265 .388 .433 .451* .463 .475* .480* .485* .485*
HCl Vin GJ Q Cof NaCl Sac Cas M Suc
d_{fis}=.062
d_{tuk}=.096
Coffee
.142 .362 .370 .432 .451* .473* .475* .480* .490* .519*
Cof Q Vin GJ HCl M Sac Cas Suc NaCl
d_{fis}=.051
d_{tuk}=.079
Quinine
.262 .347 .362 .421 .413 .433 .460 .461 .470 .487*
Q Sac Cof Vin NaCl HCl Suc Cas GJ M
d_{fis}=.062
d_{tuk}=.096
NaCl
.117 .359 .413 .439 .441 .458 .463 .465 .471 .519*
NaCl Sac Q Suc Cas GJ HCl M Vin Cof
d_{fis}=.070
d_{tuk}=.109


Sucrose
.082
.238
.365
.366
.421
.439
.460
.485*
.490*
.499*
Suc Sac GJ Cas M NaCl Q HCl Cof Vin
d_{fis}=.056
d_{tuk}=.088
Saccharin
.134
.185
.238
.347
.359
.378
.382
.463
.475*
.475*
Sac Cas Suc Q NaCl M GJ Vin Cof HCl
d_{fis}=.064
d_{tuk}=.100
Casein
.111
.185
.247
.366
.367
.441
.450
.461
.480*
.480*
Cas Sac M Suc GJ NaCl Vin Q HCl Cof
d_{fis}=.053
d_{tuk}=.083
Milk
.082
.247
.372
.378
.421
.465
.471
.473*
.485*
.487*
M Cas GJ Sac Suc NaCl Vin Cof HCl Q
d_{fis}=.056
d_{tuk}=.087
Grape Juice
.250
.365
.367
.372
.382
.388
.432
.458
.470
GJ Vin Suc Cas M Sac HCl Cof NaCl Q
d_{fis}=.064
d_{tuk}=.100

mean CPR is shown for rats tested on the flavor which is common to the entire row and trained to have an aversion to the flavor labelled on the abscissa; this is called the direct CPR. In the case of the closed bars, the mean CPR is shown for animals tested on the abscissa flavor and trained on the flavor which is common to the entire row; this called the indirect CPR. Of course, if there are two open bars or two closed bars, the left one refers to Experiment 1 and the right one refers to Experiment 2. In the case of NaCl, only three bars are presented due to the experimental error mentioned in the procedure; that is, there were no data for the rats tested on NaCl but trained on another substance. Finally, the HCl CPRs were only available in Experiment 2.

The figures show up to four measures of the generalization between two flavors. It is evident from the figures that these different measures yield similar results. Given these similar results, any possible gain in statistical sensitivity to be obtained by making a statistical distinction between whether these measures were direct or indirect or whether they were from Experiment 1 or 2 would be too small to warrant the additional statistical

complexity. Therefore, for each analysis, we used scores pooled over both Direct/Indirect CPRs and Experiments. The mean pooled CPR is presented over each abscissa flavor in Figs. 1 & 2. There were three important statistical questions to be answered as follows.

(1) The first question was whether any two flavors were independent of one another, that is whether an aversion to Flavor A showed no generalization with Flavor B. Such independence would be demonstrated if the CPRs were not reliably below 0.5 at the one - tail .05 level according to t tests. Such pairs of flavors are defined as orthogonal here and are marked with an asterick in Figures 1 and 2. As is seen, the following flavor pairs are orthogonal by our definition: Vinegar-Sucrose, HCl-Coffee, HCl-Casein, HCl-Saccharin, HCl-Milk, HCl-Sucrose, Coffee-Milk, Coffee-Casein, Coffee-Saccharin, Coffee-Sucrose, Coffee-NaCl and Quinine-Milk.

We do not claim that there is absolutely no generalization between flavors we have defined as orthogonal. Note in Figures 1 and 2 that in all but one of the 12 cases of orthogonality mentioned above, the obtained value was under 0.5, thus,

overall, their central tendency is below 0.5 ($p < 0.01$, binomial test), which means that they are not truly orthogonal. Furthermore, under different experimental conditions, Revusky, Parker, Coombes and Coombes (1976) failed to find orthogonality between vinegar and sucrose (CPR≈.439) even though the present mean CPR = .499. However, we say with confidence that there is very little generalization between these "orthogonal" flavors. Indeed, it is conceivable that the small amount of generalization which exists is due to association of the drinking response with toxicosis and not generalization of the flavor.

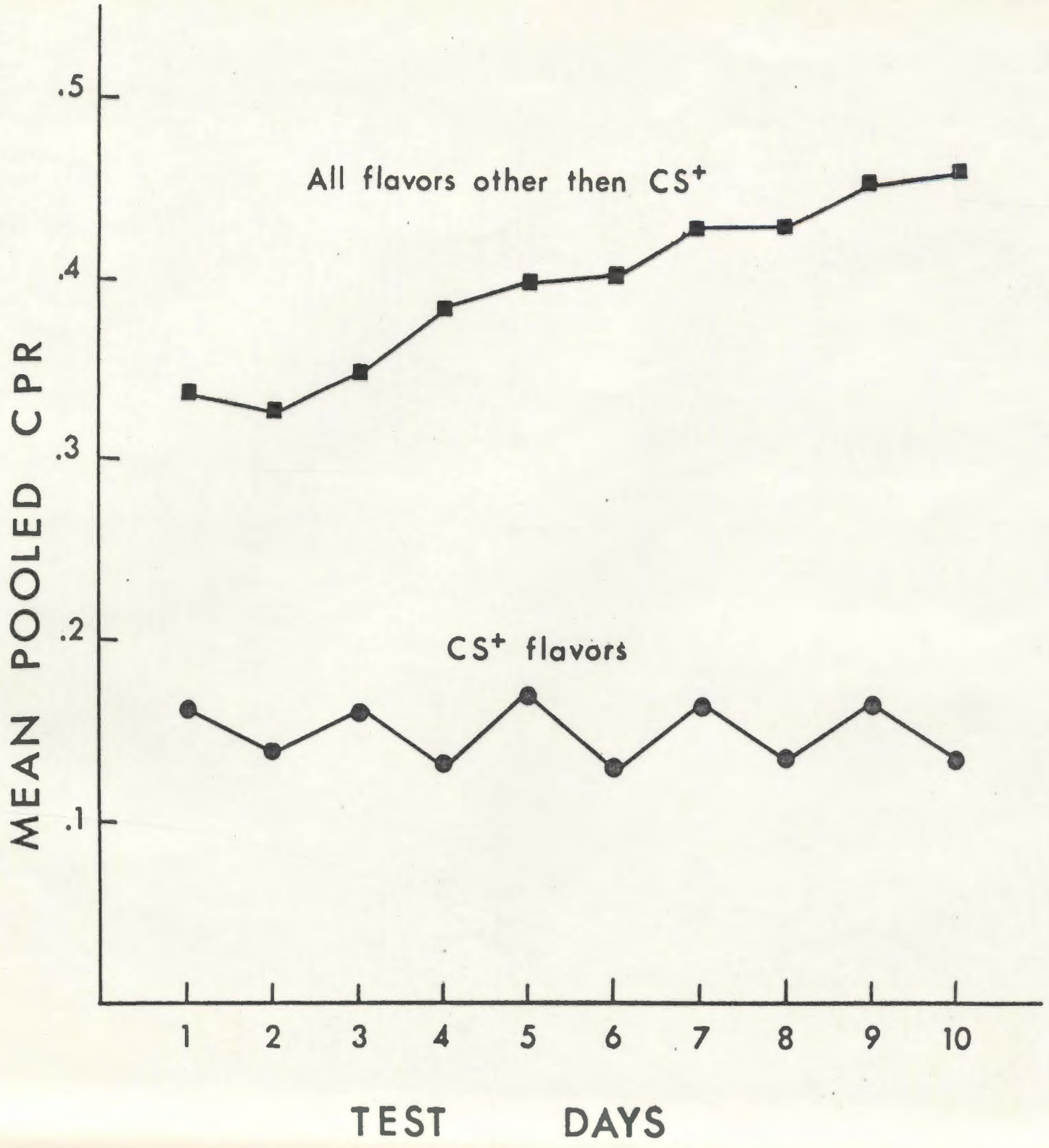
(2) The second question is whether there was complete generalization between any two flavors; the answer to this is that there was not. To assess complete generalization between flavor A and flavor B, we placed in one set CPRs for animals trained and tested on A and animals trained and tested on B; in the second set were CPRs for the generalization between A and B. We define complete generalization as the failure to obtain a significant difference ($p < .05$) between the sets of scores. In each case of such a comparison, we reached $p < .01$ by one-tailed t test.

(3) The third question was whether there were any differences in the degree with which a given flavor generalized with each of the remaining flavors. For instance, in the sucrose section of Figure 1, it is evident that casein generalized more with sucrose than did vinegar. In order to assess these differences, individual flavors, represented in each of the figures, were analyzed by an unweighted means analysis of variance. The CPRs were pooled for all cases where they involved the same two flavors, but the CPRs for which the testing and training flavors were the same was not used. In every case, $F > 5.2$, with 8 df in the numerator and 177-299 df in the denominator, $p < .001$. Using the error term for F, we have calculated the difference between mean CPRs which is necessary for a two-tailed $p < .05$ according to the t-test (based on the geometric mean of n) and called it d_{fis} as shown in Figures 1 and 2. A result significant by the d_{fis} criterion is not controlled for experimentwise error; we do not think this is necessary because of the low probabilities of the overall F's. However, we also supply d_{tuk} in the figures; if the difference between mean CPRs is greater than d_{tuk}, it is

significant according to Tukey's Honestly Significant Difference Method.

Finally, we point out results which are germane to the question of generalized extinction of the aversion over test days. 1) There was no generalized extinction of flavor aversions as a result of exposure to other flavors. To test this, we used CPRs from both experiments for cases in which the training and testing flavor was the same. The columns of the ANOVA were successive Test Days the rows were flavors. Using the interaction as the error term we did not obtain a significant Days effect ($F_{(9,100)}=0.4$) as is shown in the lower curve of Figure 3. However, there was a difference among the flavors in the strength of the aversion ($F_{(9,100)}=4.9$; $p < .01$). 2) There was a decrease in generalization between flavors over days. The CPRs for all flavors in both experiments excluding the flavor which was used in both training and testing, were pooled for each test day. A one-way ANOVA for days revealed a significant increase in CPRs over test days ($F_{(9,2636)}=19.3$; $p < .01$). The results are shown in the upper curve of Figure 3. Figure 3 suggests that the aversions to the CS+

Figure 3. Mean pooled CPR on each test day for rats which consumed their CS+ flavor (lower curve) and for rats which consumed flavors other than their CS+ flavor (upper curve).


.5
.4
.3
.2
.1
MEAN POOLED CPR
All flavors other then CS+
CS+ flavors
1
2
3
4
5
6
7
8
9
10
TEST DAYS

flavors were too strong to be weakened as a result of exposure to other than the CS+ flavor. However, the generalized aversions, being weaker, were reduced by exposure to other flavors (which included the CS+ flavor).

Discussion

As a consequence of prior toxicosis training with one flavored solution, a different novel flavor never associated with toxicosis is less likely to be consumed. Best and Batson (1977) have suggested that an aversion to a training flavor generalizes to a different test flavor because both flavors are novel to the animals; however, this "Conditioned novelty aversion" cannot explain why some novel flavors do generalize and other novel flavors do not generalize with a conditioned aversive flavor.

As is evident in Figures 1 and 2 above, each flavor had its own generalization gradient. Relatively orthogonal flavor pairs were obtained which correspond with the four primary taste categories. Sweet (Sucrose) was orthogonal with both Sour (HCl and Vinegar) and Bitter (Coffee, but not Quinine). Bitter Coffee was orthogonal with both Sour HCl (but not Vinegar, as previously noted by Siegel, 1974) and Salty (NaCl). However, there was some generalization between Sweet (Sucrose) and Salty (NaCl), a finding previously suggested by Revusky, Parker, Coombes and Coombes (1976), and between Sour (HCl and Vinegar) and

Salty (NaCl).

Stimulus generalization has been historically explained in terms of common elements shared between stimuli. Our results show that, indeed, laboratory rats, having previous experience with only the tastes inherent in rat chow and unflavored water, were capable of discriminating between elements which were shared and elements which were different across a large number of flavored solutions.

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APPENDIX C

ANOVA Summary Tables for suppression tests after Day 1 in Experiments 4, 5 and 6

TABLE 4 - 1. ANOVA Summary Table for the latency data of Experiment 4 on Days 2 and 3. A 2 x 2 ANOVA with the factors of Infusion condition and flavor of infusion.

Source of variance	df	MS	F	p
Day 2				
A (Infusion condition)	1	937.500	7.443	.01
B (Flavor of infusion)	1	88.935	.706	
AB	1	8.402	.067	
S	20	125.961		
Day 3				
A	1	237.510	3.538	.10
B	1	65.670	.978	
AB	1	222.650	3.317	
S	1	67.128		

TABLE 4-2. ANOVA Summary Table for the lick rate data on Days 2 and 3 of Experiment 4. A 2 x 6 repeated measures ANOVA with the factors of Infusion condition and 30-second intervals.

TEST DAY 2

Source of variance	df	MS	F
Between Subjects			
A (Infusion condition)	1	1527.503	.302
S	22	5062.124	
Within subjects			
B (30 sec. intervals)	5	731.805	1.047
AB	5	55.337	.078
SB	110	698.910	

TEST DAY 3

Between Subjects			
A	1	11147.901	2.578
S	22	4324.132	
Within Subjects			
B	5	770.414	1.415
AB	5	763.421	1.402
BS	110	544.506	

TABLE 5 - 1. ANOVA Summary Table for the latency data on Days 2, 3, 4 and 5 of Experiment 5. A 2 x 2 x 2 ANOVA with the factors of Infusion condition, Flavor of infusion, Flavor of Test solution.

Source of Variance	df	MS	F	p
	Day 2			
A (Infusion Condition)	1	1088000.0	30.562	.001
B (Flavor of Infusion)	1	22204.2	.624	
C (Flavor of TS)	1	130537.0	3.667	
AB	1	108003.0	3.034	
AC	1	136503.0	3.834	
BC	1	27337.6	.768	
ABC	1	24705.0	.694	
S	16	35599.6		
	Day 3			
A	1	187267.0	11.416	.01
B	1	11266.7	.687	
C	1	15000.0	.914	
AB	1	2816.8	.172	
AC	1	18150.0	1.106	
BC	1	46816.6	2.854	
ABC	1	26666.4	1.626	
S	16	16404.2		
	Day 4			
A	1	190816.0	8.770	.01
B	1	109350.0	5.026	.05
C	1	21600.0	.993	
AB	1	13066.9	.601	
AC	1	1350.1	.062	
BC	1	22816.7	1.049	
ABC	1	2399.8	.110	
S	16	21758.3		
	Day 5			
A	1	56066.6	4.482	.10
B	1	11266.7	.901	
C	1	18150.0	1.451	
AB	1	5400.1	.432	
AC	1	12149.9	.971	
BC	1	6016.7	.481	
ABC	1	2016.7	.161	
S	16	12508.3		

TABLE 5 - 2. ANOVA Summary Table for the lick rate data on Days 2, 3, 4 and 5 of Experiment 5. A 2 x 6 repeated measures ANOVA with the factors of Infusion condition and 30-second intervals.

Source of variance	df	MS	F
Day 2			
Between Subjects			
A (Infusion condition)	1	88.673	.01
S	22	8203.928	
Within Subjects			
B (30-sec.intervals)	5	113.490	.13
AB	5	534.590	.62
SB	110	864.073	
Day 3			
Between Subjects			
A	1	12432.219	.92
S	22	13529.344	
Within Subjects			
B	5	42.133	.06
AB	5	775.949	1.17
SB	110	662.651	
Day 4			
Between Subjects			
A	1	1640.250	.22
S	22	7491.520	
Within Subjects			
B	5	3457.812	2.93
AB	5	350.437	.30
SB	110	1178.810	
Day 5			
Between Subjects			
A	1	935.340	.23
S	22	3908.696	
Within Subjects			
B	5	666.690	.75
AB	5	1176.123	1.32
SB	110	890.997	

TABLE 6 - 1. ANOVA Summary Table for the latency data on Days 2, 3, 4, and 5 of Experiment 6. A 2 x 2 x 2 ANOVA with the factors of Infusion condition, Flavor of infusion, Flavor of test solution.

Source of Variance	df	MS	F	p
		Day 2		
A (Infusion Condition)	1	95886.900	6.671	.025
B (Flavor of Infusion)	1	.375	.000	
C (Flavor of TS)	1	19551.000	1.360	
AB	1	84.375	.001	
AC	1	10209.400	.710	
BC	1	13113.400	.912	
ABC	1	18095.100	1.259	
S	16	14372.800		
		Day 3		
A	1	66570.500	3.407	.10
B	1	1232.670	.063	
C	1	42841.400	2.193	
AB	1	24.000	.001	
AC	1	33004.200	1.689	
BC	1	48.148	.002	
ABC	1	661.562	.034	
S	16	19539.200		
		Day 4		
A	1	31755.300	2.107	
B	1	11310.000	.751	
C	1	13776.000	.914	
AB	1	5370.020	.356	
AC	1	14259.400	.946	
BC	1	7957.000	.528	
ABC	1	7884.440	.523	
S	16	15068.600		
		Day 5		
A	1	96.000	.223	
B	1	160.167	.372	
C	1	1380.170	3.207	
AB	1	1148.170	2.668	
AC	1	748.167	1.738	
BC	1	2242.670	5.211	.05
ABC	1	150.000	.349	
S	16	430.373		

TABLE 6 - 2. ANOVA Summary Table for the lick rate data on Days 2, 3, 4 and 5 of Experiment 6. A 2 x 6 repeated measures ANOVA with the factors of Infusion condition and 30-second intervals.

Source of variance	df	MS	F	p
Day 2				
Between Subjects				
A (Infusion condition)	1	6426.680	1.24	
S	22	5196.130		
Within Subjects				
B (30-sec. intervals)	5	96.294	.16	
AB	5	197.012	.33	
SB	110	588.737		
Day 3				
Between Subjects				
A	1	733.507	.10	
S	22	7635.977		
Within Subjects				
B	5	305.507	.31	
AB	5	476.673	.49	
SB	110	980.541		
Day 4				
Between Subjects				
A	1	330.027	.06	
S	22	5933.703		
Within Subjects				
B	5	334.861	.49	
AB	5	414.510	.61	
SB	110	682.754		
Day 5				
Between Subjects				
A	1	8680.016	1.14	
S	22	7613.910		
Within Subjects				
B	5	608.583	.76	
AB	5	297.777	.37	
SB	110	804.849		

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