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Flavor-flavor associations induce hedonic shifts in taste preference

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Two experiments allowed rats to drink freely two neutral flavors (almond and vanilla) in simultaneous compound with two hedonically valued flavors (quinine and saccharin). The neutral flavor previously paired with saccharin was subsequently preferred. The neutral flavor that had been paired with quinine was subsequently avoided. Experiment 3 found similar results when the animals were hand-fed a preset amount of the solution. Preference shifts were not obtained when differential amounts of the neutral flavors were consumed in isolation. The data indicate that flavor-flavor associations can shift taste preferences.

When a neutral tasting substance has previously been paired with a substance that produces illness, rats will refuse to drink the neutral tasting substance (see Domjan, 1980, for a review). Conversely, a neutral taste that has been paired with recovery from illness becomes preferred (e.g., Green & Garcia, 1971; Soughers & Etscorn, 1980; Zahorik, Maier, & Pies, 1974). It has been hypothesized that this change in consummatory behavior occurs because of a hedonic shift in taste quality (Garcia, Hankins, & Rusiniak, 1974). Associating a flavor with illness decreases its hedonic value; associating a flavor with an improvement in the internal milieu increases its hedonic value. That is, conditioning produces a shift in palatability that influences consumption on the test trial.

Flavors, of course, are not hedonically neutral to begin with. The appropriate combination of saccharin and water has positive hedonic value (it is preferentially consumed over water), while a solution of quinine and water has negative hedonic value (it is rejected). It seems plausible that if a flavor with strong hedonic value (such as saccharin or quinine) is paired with a flavor of neutral hedonic value (such as vanilla), the former might cause a shift in the hedonic value of the latter. That is, if we establish an association between two flavors, we might change the palatibility of one of these flavors.

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There is good evidence that associations can be formed between two flavors (Lavin, 1976; Rescorla & Cunningham, 1978). However, this research has typically been carried out in a framework of sensory preconditioning. Two putatively neutral flavors (S1 and S2) are paired, and then one (S1) is subsequently paired with illness. The flavor-flavor association is then indicated by a shift in preference or consumption of the other flavor (S2). This paradigm indicates that flavor-flavor associations occur, although it does not provide evidence that a flavor-flavor association in and of itself can produce a hedonic shift.

Bolles, Hayward, and Crandall (1981) have demonstrated that rats will show a conditioned preference for a neutral flavor (almond, anise, or vanilla) after the flavor has been paired with a diet high in starch content. They suggest that the taste of starch might cause this positive hedonic shift in the neutral flavor. However, the purpose of Bolles et al.'s experiment was not to demonstrate that such flavor-flavor associations induced hedonic shifts, and thus their data remain only suggestive. For example, the caloric consequences of starch may be necessary to produce this preference. Also, it has not been demonstrated conclusively that starch possesses positive hedonic palatability (and the diets used by Bolles et al. were not markedly different in taste).

There is evidence that a flavor paired with saccharin may become preferred to one that has not been paired with saccharin (Holman, 1980) or to one that has been paired with a more dilute saccharin solution (Holman, 1975). However, the converse, that a neutral flavor experienced in compound with an aversive taste will subsequently be avoided, has yet to be demonstrated.

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Thus, the purpose of the present experiment was to demonstrate hedonic shifts due exclusively to flavor-flavor associations. We also tried to determine if we could induce a positive and a negative shift with appropriate hedonically valued flavors. If a neutral flavor, such as vanilla, has been paired with a palatable saccharin solution, will vanilla be preferred? Likewise, if vanilla is paired with an aversive tasting quinine solution, will vanilla be rejected?

EXPERIMENT 1

Method

Subjects. Ten female Long-Evans-strain rats (Blue Spruce Farms; Altamont, N.Y.) were used. They had been used previously in a fear conditioning experiment but had had no experience with any deprivation schedules. They were approximately 180 days old and weighed 220-300 g. The animals were individually housed in standard cages with sheet-metal sides and a wire-mesh bottom. The cages were kept in a colony room maintained on a reverse light-dark cycle (12 h:12 h; lights off at 1000 h). Before the experiment started, the animals had free access to Purina Rat Chow and tap water.

Materials. Durkee's imitation vanilla and pure almond extracts were used. Quinine was obtained from Sigma Chemical Corp. (St. Louis, Mo.); the saccharin, in effervescent tablet form was obtained at a local drug store. Solutions were presented to the animals in glass bottles with ball bearing sip tubes. Consumption was determined by weighing the bottles both before and after presentation. The saccharin solutions contained .2% (wt/vol) sodium saccharin, and the quinine solutions contained .01% (wt/vol) quinine sulfate in tap water at room temperature. Vanilla- and almond-flavored solutions contained 4% (vol/vol) of these substances. Actually six solutions were used: a vanilla flavored saccharin, a vanilla flavored quinine, an almond flavored saccharin, an almond flavored quinine, vanilla alone, and almond alone.

Procedure. Animals stayed in their home cages throughout the experiment. The experiment began by adapting the animals, for 3 days, to a 23-h deprivation schedule. The animals were allowed daily access to water, for 1 h at 1600 h. Two bottles, each containing tap water at room temperature, were presented simultaneously for that hour. Food was removed from the cages just before water was given and was returned immediately after the bottle was removed. On Day 4, the animals were divided into two equally sized groups for conditioning. Conditioning occurred on Days 4-9. On even-numbered days, Group VS/AQ received two bottles, both containing a vanilla-saccharin solution, while Group AS/VQ received two bottles, both containing an almond-saccharin solution. On odd-numbered days, Group VS/AQ received an almond-quinine solution in both bottles and Group AS/VQ received a vanilla-quinine solution in both bottles. Except for the bottle's contents, the conditioning procedure was like that of the adaptation procedure. So, Group VS/AQ had vanilla paired with saccharin and almond paired with quinine; Group AS/VQ had the reverse combination.

Days 10 and 11 were test days. On both days, each group was presented one bottle of vanilla alone and one bottle of almond alone, and allowed to make a free choice between these solutions. Otherwise, the test procedure was similar to the conditioning and adaptation procedure. The position of the flavors was reversed between test days.

The data were analyzed as a preference ratio for vanilla: the amount of vanilla consumed divided by the total amount of the two flavors consumed. A ratio greater than .5 indicates a preference for vanilla relative to almond. A ratio below .5 indicates an aversion to vanilla relative to almond. A ratio of .5 indicates equal preference for the two flavors.

Results and Discussion

During the test days, every animal in the group that had vanilla paired with saccharin and almond paired with quinine (Group VS/AQ) preferred vanilla (mean $\pm$ SEM preference ratio on Day 10 = .92 $\pm$ .01 and on Day 11 = .86 $\pm$ .07). Every animal in the group that had vanilla paired with quinine and almond paired with saccharin (Group AS/VQ) found vanilla aversive (mean $\pm$ SEM preference ratio on Day 10 = .15 $\pm$ .09 and on Day 11 = .22 $\pm$ .09). The differences between these means were reliable for both test days [$ts(8) > 5.81$, $p < .01$]. There was no overlap in the scores of the two groups on either test day [$Us(5,5) = 0$, $p < .01$]. During conditioning, the rats drank 15.7 $\pm$ .9 (mean difference $\pm$ SEM) more of the saccharin-containing solution than of the quinine-containing solution [$t(9) = 17.38$, $p < .01$]. This difference indicates that the rats preferred saccharin to quinine. The test days' data indicate that rats prefer a flavor that has a previous history of being in compound with a hedonically positive flavor over one that has a history of being in compound with a hedonically negative flavor.

EXPERIMENT 2

The hedonically valued flavors did induce a shift in preference to the more neutral flavors. However, the design of the experiment did not allow us to distinguish whether quinine conditioned an aversion, if saccharin conditioned a preference, or if both occurred. The design of Experiment 2 allowed us to assess both a preference for one flavor and an aversion to the other in reference to a control group that should show neither a preference nor an aversion.

Method

Subjects and Materials. Fifteen rats, similar to those of the first experiment, were used. Housing conditions and materials were those of the first experiment.

Procedure. Three groups of five animals each were adapted to the same watering and feeding schedule as in Experiment 1 for 3 days. Water was given at 1400 h. Days 4-15 were conditioning days, and for each rat both bottles contained the same solution. The solutions that each group was exposed to are presented in Table 1. Group VS/AQ was designed to establish associations of vanilla with saccharin and almond with quinine. Group AS/VQ was designed to produce associations of almond with saccharin and vanilla with quinine. Group CE was a control that had exposure to quinine, saccharin, almond, and vanilla equivalent to that of the experimental groups, but in a manner designed not to foster any associations.

Days 16 and 17 were test days. On Day 16, almond alone was tested: all groups had a choice between almond-flavored tap water in one bottle and plain tap water in the other. Day 17 was a vanilla test day: all groups had vanilla-flavored tap water in one bottle and plain tap water in the other.

The data for these test days was a preference ratio for the flavored solution. This ratio was equal to the amount of the flavored solution consumed divided by the total amount of both solutions consumed during the test. All other aspects of the procedure were as they were for Experiment 1.

Results and Discussion

The preference ratio data are summarized in Table 1. The first test day's data (almond test) and the second test day's data (vanilla test) were each analyzed separately by a one-way analysis of variance.

The overall ANOVA on the first test day's data was reliable [$F(2,12) = 34.17$, $p < .001$]. In comparison with controls (Group CE), animals that had had almond paired with quinine (Group VS/AQ) had an *aversion* to almond alone [$F(1,12) = 30.64$, $p < .001$] and animals that had had almond paired with saccharin (Group AS/VQ) had a *preference* for almond alone [$F(1,12) = 6.56$, $p < .025$]. Thus, quinine conditioned an aversion to almond while saccharin conditioned a preference for almond.

The results of the second test day were less clear. Neither the overall ANOVA [$F(2,12) = 2.09$] nor comparisons of the two experimental groups (Groups VS/AQ and AS/VQ) with the control group (Group CE) were reliable [$Fs(1,12) \leq 1.25$]. However, animals that had had vanilla paired with saccharin (Group VS/AQ) had preference ratios that were marginally larger than those of the animals that had had vanilla paired with quinine (Group AS/VQ), indicating that there was a trend in the same direction as that obtained on the first test day [$F(1,12) = 4.27$, $p = .06$].

There are several possible reasons why vanilla did not condition as successfully as almond. First, it is possible that almond is a better or more salient CS. Second, rats seemed to prefer vanilla to almond. The controls, which drank both vanilla alone and almond alone during conditioning, drank, on the average, 2 ml/h more vanilla than almond during conditioning, and this difference was reliable [$t(4) = 3.48$, $p < .02$]. Possibly this bias toward vanilla's taste offset the conditioned changes in vanilla's taste quality. A third possibility is that the prior test with almond somehow diminished the conditioned changes in the taste quality of vanilla.

During conditioning, Groups VS/AQ and AS/VQ had a mean daily intake of 19.6, 3.8, and 17.7 g of the saccharin-flavored compound, quinine-flavored compound, and water, respectively. A repeated measures analysis of variance showed that these means were different [$F(2,18) = 142.86$, $p < .001$]. The rats drank reliably more water than quinine [$F(1,18) = 201.26$, $p < .001$] and marginally more saccharin than water [$F(1,18) = 3.76$, $p = .068$]. While these differences support our assumption that quinine is aversive and saccharin is pleasant, they also indicate that there is a confound present in Experiments 1 and 2; there was differential consumption of the quinine-paired and saccharin-paired flavors. The shift in preferences that we found may not be due to an association between the neutral flavor and the hedonically valued flavor, but to the differential consumption of the neutral flavors during conditioning. Experiments 3, 4, and 5 tested these two alternatives. Experiment 3 determined if quinine and saccharin could shift the preferences for neutral flavors in the absence of intake differences. Experiments 4 and 5 determined if shifts in preferences for the neutral flavors could be caused merely by differential amounts of consumption.

EXPERIMENT 3

Experiment 3 was essentially a replication of the first experiment with modifications that allowed us to control the animals' intake and insure equal consumption of both the saccharin- and quinine-containing compounds. Rather than freely drinking from water bottles, the animals were hand-fed predetermined amounts of the compound flavors.

Method

Subjects and Apparatus. Sixteen animals like those of the first experiment served. Although housing conditions were similar in both experiments, these animals were run during the lights-on portion of the light-dark cycle. An ample supply of food was maintained continuously in the home cage.

The same solutions as in the first experiment were used, except that vanilla concentration was increased to 8% (vol/vol). The rats were fed the solutions via a 5-cc hypodermic syringe (B-D Plastipak). The syringe guard cap was left on the syringe and a 1.5-mm-diam hole was punched into its tip.

Table 1
Solutions Presented to the Three Groups on the 17 Days of Experiment 2

	Day																		Test Results			
	Adaptation			Conditioning													Test With Water		Day 16*		Day 17†	
Group	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	Mean	SEM	Mean	SEM
VS/AQ	W	W	W	V-S	A-Q	W	W	V-S	A-Q	W	W	V-S	A-Q	W	W	A vs. W	V vs. W	.07	.02	.69	.10
AS/VQ	W	W	W	A-S	V-Q	W	W	A-S	V-Q	W	W	A-S	V-Q	W	W	A vs. W	V vs. W	.70	.07	.30	.16
CE	W	W	W	A	V	Q	S	V	A	Q	S	A	V	Q	S	A vs. W	V vs. W	.27	.06	.51	.14

*Note—W = plain water; A = almond water; V = vanilla water; S = saccharin water; Q = quinine water; A-S = almond-flavored quinine water; V-Q = vanilla-flavored quinine water. Each animal was presented with two bottles simultaneously. On Days 1-15, the two bottles contained the same solution. On Days 16 and 17, the two bottles contained different solutions. The test results are the mean preference ratios for the test fluid. *Almond. †Vanilla.*

Procedure. The following procedure was followed for 11 days: A rat was removed from its home cage and held against the experimenter's chest. The experimenter then fed the animal 3.5 cc of fluid. The plunger was depressed in a slow and continuous manner so that it took 90 sec to empty the syringe. If the rat would not voluntarily lick the fluid, the tip of the syringe guard was gently forced behind the rat's upper front teeth. Fluid infusion invariably elicited licking movements when the syringe was positioned in such a manner. Fluid was released from the syringe only when the rat was licking or when the syringe opening was inside the rat's mouth. It took about 2 min to feed a rat in this manner.

Fifteen minutes after hand-feeding, the rat was given 12 min of access to a water bottle containing tap water. The rat received no other fluid. All rats received water from the syringe for the first 5 days. The eight animals in Group AS/VQ were hand-fed an almond-saccharin compound on Days 6, 8, and 10 and a vanilla-quinine compound on Days 7, 9, and 11. The eight animals in Group AQ/VS were hand-fed an almond-quinine compound on Days 6, 8, and 10 and a vanilla-saccharin compound on Days 7, 9, and 11.

Day 12 was a test day. No hand-feeding occurred on this day. All the rats received free access to one bottle of almond alone and one bottle of vanilla alone. The bottles were presented simultaneously and their positions were switched after 6 min. After 12 min, the bottles were removed and weighed. The dependent variable was the preference ratio for vanilla obtained on the test day.

Results and Discussion

By the fifth water day, all the rats readily licked the fluid from the syringe. On the first compound day (Day 6), the animals in Group AS/VQ readily drank their almond-saccharin. While Group AQ/VS readily took a few initial licks of their almond-quinine, they had to be force-fed the remainder. On Day 7, Group AS/VQ voluntarily took a few initial licks of vanilla-quinine but had to be force-fed the remainder. Group AQ/VS initially had to be forced to take vanilla-saccharin but voluntarily drank approximately the last 2 cc. From that point on (Days 8-11), all animals refused the quinine-containing solution after the first few licks but readily drank the saccharin-containing solution, usually after being forced to drink the initial .5 cc or so.

All animals in Group AQ/VS showed a preference for vanilla on the test day (the mean ± SEM preference ratio was .81±.02). Six of eight animals in Group AS/VQ showed an aversion to vanilla (.41 ±.07). This difference was reliable [$t(14) = 5.32$, $p < .01$], and there was little overlap in the distribution of scores [Mann-Whitney $U(8,8) = 1$, $p < .01$]. We found that, even when amount of consumption is equated, rats prefer a flavor that has a history of being in compound with a hedonically positive flavor over one that has a history of being in compound with a hedonically negative flavor.

There is a potential confound in this experiment. Rats had to be force-fed more of the quinine-containing compound, while they voluntarily consumed more of the saccharin-containing compound. Possibly, force-feeding is aversive and the preference shift was induced, not by the hedonic value of the flavor, but by the differential amounts of force-feeding. This explanation cannot account for the results of the first two experiments, but they can be accounted for by differential consumption. Therefore, Experiments 4 and 5 determined if differential consumption could cause a shift in taste preference.

EXPERIMENT 4

In this experiment, consumption of almond alone and vanilla alone was varied by limiting access to these solutions. However, total intake was held constant by giving water supplements. The animals received no exposure to quinine or saccharin. If a difference in the amount of the neutral target flavor consumed was responsible for the shift in taste preferences, then this procedure should also produce a shift in taste preferences.

Method

Subjects and Materials. The subjects were 24 rats like those used in Experiment 1. They were housed under conditions like those described in Experiment 3. Two solutions were used: an 8% vanilla and tap water solution and a 4% almond and tap water one. Solutions were at room temperature.

Procedure. The rats were divided into two groups: Group A2/V10 and A10/V2. The letters indicate the nature of the solution, and the numbers indicate the number of minutes of exposure to that solution. The animals received 12 min access to fluid each day for 11 days. Both groups received water for the first 5 days. Days 6-11 were conditioning days. Each animal received exposure to one of the flavors daily. The flavors alternated daily. Half of the rats in each group received almond on Day 6 and the other half received vanilla. If the animal was scheduled to receive a 2-min exposure to a flavor, then a bottle of water was presented for 10 min immediately upon removal of the flavor. If the animal was scheduled to receive a 10-min exposure to a flavor, then water was provided for 2 min immediately upon removal of the flavor. Day 12 was the test day; it was a replication of the 12-min test procedure of Experiment 3.

Results and Discussion

The data for conditioning and testing are presented at the top of Table 2. As averaged over the 6 conditioning days, the rats drank 8.1 g more of the 10-min-exposure flavor than of the 2-min-exposure flavor. A correlated sample t test showed this difference to be reliable [$t(23) = 15.55$, $p < .001$]. This procedure was successful in producing differential consumption of the two flavors.

During conditioning, the rats in Group V2/A10 drank less almond than Group V10/A2 drank vanilla [$t(22) = 2.27$, $p < .02$], indicating a possible preference for vanilla over almond. The difference in consumption between almond and vanilla during brief exposures was not reliable [$t(22) = 1.57$], possibly due to a floor effect.

The data of principal interest are those of the test day. If differential consumption was responsible for the effects found in Experiments 1 and 2, we would expect Group V10/A2 to have a greater peference for vanilla than Group V2/A10. While the difference in

Table 2

Group	Amount Consumed During Conditioning: Vanilla Mean	Vanilla SEM	Almond Mean	Almond SEM	Preference Ratio* Mean	Preference Ratio* SEM
			Experiment 4			
V2/A10	5.3	.3	12.3	.5	.65	.05
V10/A2	14.0	.6	4.7	.3	.58	.06
			Experiment 5			
V3/A12	4.5	.4	11.4	.6	.58	.08
V12/A3	13.4	.4	4.0	.4	.48	.07

**For vanilla during testing.*

preference ratios for the two groups was not reliable [$t(22) = .8$], the trend was in the opposite direction from that predicted by the differential consumption account.

EXPERIMENT 5

In Experiment 4, consumption of the flavors was manipulated while total consumption was held constant through the use of water supplements. In Experiments 1 and 2, when consumption was reduced by quinine, no water supplements were provided and deprivation level may have increased following exposure to the quinine compound. If this was the case, the saccharin-paired flavor may have been consumed under greater deprivation conditions than the quinine-paired flavor. Revusky (1968) has shown that drinking a flavor in the presence of greater deprivation can produce a shift in flavor preference. To test the possibilty that the preference shifts in Experiments 1 and 2 were produced by consumption in the presence of differing deprivation levels, Experiment 4 was replicated but without providing water supplements.

Method

Subjects and Materials. Twenty-four rats like those of Experiment 1 were used. They were housed under the conditions described for Experiment 3. The materials were those of Experiment 4.

Procedure. The procedure was like that of Experiment 4, with the following exceptions: The rats were divided into two groups of 12. Group V3/A12 had 3-min access to vanilla alone and 12-min access to almond alone on alternating days. Group V12/A3 had 12-min access to vanilla alone and 3-min access to almond alone on alternating days. No water supplements were given.

Results and Discussion

During the course of the experiment, one animal in Group V3/A12 became severely dehydrated and too weak to drink. This animal was removed from the experiment and data from just the remaining 11 animals in Group V3/A12 are presented.

The data for conditioning and testing are presented at the bottom of Table 2. During conditioning, the rats drank, on the average, 8 g more per day of the flavor exposed for 12 min than they did of the flavor exposed for 3 min. This difference is reliable [$t(22) = 18.96$, $p < .001$]. Less almond than vanilla was drunk during 12-min exposures [$t(21) = 2.18$, $p < .05$], but the difference was not reliable for the 3-min exposures [$t(21) = .94$].

The differential consumption did not produce a reliable difference in taste preference [$t(21) = .94$]. The trend was in the unexpected direction; the rats with the shorter exposure to vanilla showed a slightly higher preference for vanilla (see Table 2).

These data indicate that consumption of the neutral flavor in the presence of differential deprivation levels cannot account for the findings of the first two experiments. We are in no way trying to say that consumption in the presence of differing deprivation levels cannot produce a shift in flavor preference; indeed, Revusky (1968) has presented convincing evidence for such an effect. We are only trying to say that such an effect is not responsible for the data reported in this series of experiments.

GENERAL DISCUSSION

Experiments 1 and 2 indicated that giving rats free access to one target flavor in compound with quinine and another target flavor in compound with saccharin will shift their preferences for the target flavors. This shift might be due either to differential amounts of consumption of the target flavors or to an association formed between the target flavors and the hedonically affective flavors they were compounded with. Our findings that a shift in preference occurs when consumption of both compounds is equated (Experiment 3) but that no preference shift occurs when different amounts of the target flavors are consumed in isolation (Experiments 4 and 5) supports the latter alternative.

Taken as a whole, the data indicate that pairing a relatively neutral flavor with another flavor that possesses hedonic taste qualities may induce a hedonic shift in the more neutral flavor. These shifts seem to be symmetrical—we can obtain them in both a positive and a negative direction (Experiment 2). Like Holman (1975, 1980), we find that pairing a flavor with a palatable saccharin solution produces a preference for that flavor. We also find that pairing a flavor with an aversive quinine solution causes that flavor alone to be rejected. Hedonic shifts can be caused by associations formed entirely in the mouth. Bolles et al. (1981) have indicated how such associations may be utilized in diet selection.

As pointed out earlier, flavor-flavor associations have been examined in the context of sensory preconditioning. Theoretically, sensory preconditioning is a demonstration of an association between two neutral stimuli (i.e., stimuli that do not condition a

CR in their one right). While, in terms of procedure, the flavor-flavor conditioning studies are similar to sensory preconditioning, theoretically they cannot qualify as demonstrations of sensory preconditioning because the present experiment shows that the putatively neutral stimuli used can actually condition a response. In this regard, our data have shown that even almond and vanilla are not entirely neutral (vanilla consumption is greater than almond consumption, Experiments 2, 4, and 5). Thus, even these flavors might have the ability to condition preference shifts.

A series of experiments within this flavor-flavor sensory preconditioning framework (Durlach & Rescorla, 1980; Rescorla & Cunningham, 1978) have interpreted performance changes due to manipulation of flavor-flavor associations in terms of the CR conditioned by a toxic US. However, we must now also consider the possibility that any manipulation of a flavor-flavor association may change performance to one of the flavors in its own right (i.e., the manipulation may affect the CR conditioned by the flavor US rather than, or in addition to, the CR conditioned by the toxic US).

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