Amount of different flavors consumed during the CS-US interval in taste-aversion learning and interference*

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Animals develop an aversion to flavors that have toxic consequences. Events occurring during the delay between the administration of the flavor and the onset of sickness are known to influence the degree of aversion. In the present study, aversion was obtained to a novel flavor, sucrose, despite the introduction of three other novel solutions (NaCl, vinegar, and coffee, presented in that order) during a 40-min delay to lithium-chloride toxicosis. When the test stimulus was coffee instead of sucrose and the amounts of consumption of the interfering flavors (vinegar, NaCl, and sucrose, presented in that order) were equated, a lesser degree of aversion was obtained, compared to the condition in which interfering solutions were allowed to be consumed in greater quantities. Aversion was not demonstrated in the latter case. In all conditions, aversions were obtained to the flavors closest in time to sickness, independent of their taste quality and the amounts of consumption of the preceding flavors. In general, coffee tended to be better interfered with by the three novel flavors, rather than with sucrose. Relatively greater consumption of the interfering flavors tended to produce greater interference, but this was thought to be interacting with the taste quality of the flavors involved.

Animals develop an aversion to flavors that have toxic consequences (Garcia, Kimeldorf, & Koelling, 1955; Revusky & Bedarf, 1967). In this paradigm, an animal is fed a nonnutritive substance, such as saccharin, and is later subjected to a mild toxicosis by such means as injection of poison-like lithium chloride or x-irradiation. After recovering from the sickness, the animals avoid the flavored solution.

Taste-aversion learning has been demonstrated to occur with one pairing and over long delays of the CS-US interval (Garcia, Ervin, & Koelling, 1966; Nachman, 1970; Revusky, 1968; Smith & Roll, 1967; Rozin & Ree, 1972). Several explanatory models have been offered in an attempt to explain this long-delay phenomenon. It has been argued that the associative strength of a stimulus with a consequence depends, in part, on the relevance of the stimulus to the consequence (Garcia & Koelling, 1966; Garcia & Ervin, 1966; Dietz & Capreta, 1967). Perhaps long delays in taste-aversion learning are possible because of the associative strength and relevance of toxicosis to ingestin.

Revusky (1971) argues that, although the learning of flavor aversions may follow the rules of the traditional associative model, the unique characteristic of long delays may be explained in terms of the lack of relevant interfering cues (other tastes) between the prior event of ingestion and the gastrointestinal consequence of sickness. This interpretation brings about the assumption

that, theoretically, if there are no relevant interfering stimuli, the delay period can be extended indefinitely. Practically, this does not seem to be the case. A number of studies have shown that increases in the delay cause decreases in learned aversion (Kalat & Rozin, 1971; Revusky, 1968; Smith & Roll, 1967).

Kalat and Rozin (1971) do not accept interference of relevant events as the sole explanation of long-delay learning. They offer the model of slow, "active" decay process whereby the "safety" of the CS is being learned. Kalat and Rozin (1971) obtained a gradient of delay when no taste was available during the CS-US interval. Furthermore, even after introducing three novel substances during the delay interval, aversion to a sucrose test stimulus was obtained. However, there was a considerable decrease in the magnitude of the aversion, suggesting that some interference might have taken place. In the Kalat and Rozin (1971) experiment, four tests were presented sequentially in an interval of 30 min. Sucrose was always the test solution and was presented either first or second after casein hydrolysate. The other substances were NaCl and coffee. The quantity of the consumed substances were not controlled.

It is a well-documented fact that stimulus magnitude and temporal contiguity are important parameters in learning (Kimble, 1961). Therefore, in the first experiment, an attempt was made to replicate the Kalat and Rozin (1971) finding of taste-aversion learning in spite of the interference of several novel flavors during the CS-US interval, with an additional control of equating the amounts of the consumed flavors. The second experiment was run to see if greater consumption

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Fig. 1. Mean volume of different flavors consumed by the TIME group over a 2½-min exposure to each during treatment in Experiment I. The constant volume of different flavors consumed by the QUANTITY group is indicated by the dotted line.

of interfering flavors would preclude aversion learning altogether.

EXPERIMENT I

Method

Subjects. Thirty female rats, weighing 100-150 g and derived from the Sprague-Dawley strain of the animal house of the American University of Beirut, Lebanon, were used as Ss.

Procedure. Rats were housed in individual mesh-wire cages. Two days of acclimation period were allowed when food (Vitasni, lab chow) and tap water were made available ad lib. Tap water was presented in two bottles. One rat died during acclimation. On Day 3, rats were randomly assigned to three groups. Pretraining commenced to familiarize them with the treatment drinking procedures, except that all the solutions were tap water. Pretraining went on for five sessions under a 47-h water-deprivation schedule.

The treatment procedures were as follows: A group of 10 rats, the QUANTITY group, were administered sucrose, NaCl, vinegar, and coffee solutions (in that order) in equal 4-ml quantities. There were 10-min intervals between the presentations of each flavor. Each solution was presented for 7½ min, which was enough time for the consumption of the available solutions. There was a 2½-min delay between the removal of one flavor and the presentation of the next. After the coffee consumption, there was a 2½-min delay to toxicosis. Another group of 10 rats, the TIME group, was presented with the same solutions, in the same order, in unlimited amounts, but for a constant duration of 2½ min only. There was a delay of 7½ min between the removal of one flavor and the presentation of another. After the presentation of the coffee, there was a 7½-min delay to toxicosis. In a third group of 9 rats, the CONTROL group, 4 ml of sucrose was presented for 7½ min and then there was a delay of 6½ h to toxicosis. Two hours before the administration of the test stimulus, 12 ml of tap water was made available for 30 min.

The flavors were solutions of 10% (W/V) sucrose; 0.15 M NaCl; 3% (V/V) Heinz cider vinegar; and 1% (W/W) Maxwell House instant coffee. Sucrose was the test stimulus in all cases. The overall delay between exposure to sucrose and toxicosis was 40 min for the QUANTITY and TIME groups. The 6½-h delay in the CONTROL group has been shown not to produce learned aversion to sucrose (Kalat & Rozin, 1971). This offers a control for familiarization with sucrose and unconditioned effects of the poisoning substance. Toxicosis was induced by IP injections of 20 ml/kg of 0.15 M lithium-chloride solution.

Forty-seven hours after the treatment, a 20-min, two-bottle preference test for sucrose was given. One bottle contained ordinary tap water and the other a concentration of sucrose equal to that used during treatment. The position of the bottles were counterbalanced within each group. Each rat was made to take several licks at both bottles before they were inserted into the cages. Forty-seven hours after the sucrose preference test, coffee (the last stimulus) aversion was measured in a 20-min one-bottle test. A one-bottle test makes the animal drink some of the unpreferred solution and thus prevents floor effect.

Results

During the treatment, flavors were consumed in different amounts. Figure 1 represents the mean volume of the different flavors consumed by the TIME group over a 2½-min exposure to each. Analysis of variance indicated significant differences in the consumptions, $F(3,27) = 6.89$, $p < .005$, and the t test for differences among several means (Bruning & Kintz, 1968) showed that sucrose was consumed significantly more than NaCl ($p < .05$), vinegar, and coffee ($ps < .01$). The QUANTITY group consumed all of the provided solutions of each of the flavors.

Table 1 presents the median consumption of sucrose (the first flavor) and water by the three groups in Experiment I. The Mann-Whitney U test was carried out (Siegel, 1956) comparing the median sucrose intake of the groups. The TIME group and the QUANTITY group were significantly less than the CONTROL group ($U = 10.00$, $p < .02$, and $U = 13.50$, $p < .02$, respectively). The comparison of the QUANTITY and the TIME groups did not yield significance, although a trend of lesser aversion was apparent in the QUANTITY group.

Table 1 also shows the median consumption of coffee (the last flavor), during the second testing session, by the three groups. The Mann-Whitney U test indicated that the TIME and QUANTITY groups had consumed significantly less coffee than the CONTROL group ($U = 6.0$, $p < .001$, and $U = 7.5$, $p < .001$, respectively), and were not different from each other.

Discussion

Experiment I successfully replicated the Kalat and Rozin (1971) finding of significant aversion to sucrose, despite interference from three novel flavors. Both conditions of stimulus presentation showed significant

Table 1
Results of Two-Bottle Sucrose Test and One-Bottle Coffee Test in Experiment I

Group	Sucrose (First Flavor) Test		Coffee (Last Flavor) Test
	Median Volume (mL) Sucrose	Water	Median Volume (mL) Coffee
Quantity	3.6	6.8	5.4
Time	1.4	10.0	4.8
Control	12.8	3.6	12.0

aversions. However, to interpret these findings as aversion-acquired, in spite of the ingestion of three novel interfering substances without regard to the relative amounts of consumption of the test and the interfering stimuli, may be premature. Examination of Fig. 1 indicates that the sucrose intake of the TIME group, on the average, is the same as that of the QUANTITY group, while the relative intake of the interfering flavors were different.

As expected, there was very strong aversion to coffee (the last flavor) by both groups. This effect may be attributed to the temporal proximity of coffee to the onset of sickness. In spite of the fact that the flavors consumed before coffee may have had interfering effects (Revusky, 1970), learned aversion to coffee was demonstrated. It should be noted also that preference for coffee was tested 4 days after treatment and 47 h after the test for sucrose preference.

EXPERIMENT II

In the first experiment, the preference for sucrose by the QUANTITY group was indicative of the greater interfering effect of the three flavors consumed in constant amounts during the delay between the sucrose intake and the administration of toxicosis. The second experiment was designed to maximize the interfering effects. In order to do this, the sequence of the flavors given during the treatment in Experiment I was reversed so that the preferred substances being presented later would be consumed in greater amounts than the earlier flavors, and an unpreferred flavor (coffee) would become the test stimulus.

Method

Subjects. Thirty-two rats from the Sprague-Dawley strain, 50-100 g, obtained from the Animal Suppliers, London, were used as Ss. One animal from the QUANTITY group and one from the CONTROL group died before the testing session. There were 9, 10, and 11 rats in the QUANTITY, TIME, and CONTROL groups, respectively.

Procedure. The procedure was similar to that of Experiment I for all three groups, with the following differences: The order of presentation of the flavors was reversed and coffee became the first, or the test, stimulus; the groups were matched by sex as well as by weight; a 23-h water-deprivation schedule was followed instead of a 47-h schedule; since the animals in this experiment were smaller, the amounts of the fluids were adjusted such that the CONTROL group drank 7½ ml (instead of 12 ml) 2 h before the treatment and 2½ ml (instead of 4 ml) of the test stimulus during treatment, and the QUANTITY group was given 2½ ml (instead of 4 ml), at a time, of a particular flavor; forty-seven hours after treatment (to allow for recovery from toxicosis) a one-bottle test (instead of a two-bottle test) for coffee preference was given to the groups. On the following day, preference for the last flavor (sucrose) was measured by another 20-min one-bottle test. The duration of exposure to a particular flavor for the TIME group was again 2½ min long.

Results

Figure 2 presents the mean volume of the different flavors consumed by the TIME group over 2½-min

Fig. 2. Mean volume of different flavors consumed by the TIME group over a 2½-min exposure to each during treatment in Experiment II. The constant volume of different flavors consumed by the QUANTITY group is indicated by the dotted line.

exposure to each during treatment. Analysis of variance showed significant differences between the mean consumption of different flavors, $F(3,27) = 18.63$, $p < .001$, and the t test for differences among several means showed that coffee was consumed significantly more than vinegar ($p < .01$), but significantly less than NaCl ($p < .05$) and sucrose ($p < .01$). The QUANTITY group consumed all of the provided solutions of each of the flavors.

Table 2 presents the median coffee consumption of three groups during the testing session. The Mann-Whitney U test revealed that the median of the QUANTITY group was less than that of the TIME group ($U = 23.00$, $p < .10$) but not different from the median of the CONTROL group.¹ The medians of the TIME and CONTROL groups were not different from each other.

Table 2 indicates also the median consumption volume of sucrose (the last flavor) by the three groups. The Mann-Whitney U test indicated that the medians of the TIME group and the QUANTITY group were less than the CONTROL group's ($U = 30.50$, $p < .10$, and $U = 14.00$, $p < .02$, respectively), and not different from each other.

Discussion

In the second experiment, no aversion was

Table 2
Results of One-Bottle Coffee Test and One-Bottle Sucrose Test in Experiment II

Group	Coffee (First Flavor Test)	Sucrose (Last Flavor) Test
	Median Volume (mL)	Median Volume (mL)
Quantity	2.4	6.4
Time	4.2	7.0
Control	4.4	9.6

demonstrated by either of the groups, compared to the CONTROL group. However, the aversion demonstrated by the QUANTITY group was greater, compared to the TIME group. This may suggest the possibility that the greater consumption of the interfering stimuli by the TIME group may have interfered to a greater extent than it did in the case of the QUANTITY group, where the test stimulus and the interfering stimuli were consumed in equal amounts.

As in the first experiment, aversion to the flavor consumed last (sucrose) was obtained. Here too, aversion was demonstrated despite the probability of extinction over time and interference from flavors consumed earlier.

Taken together, these data indicate that learning does take place in spite of large amounts of interference even when the degree of interference was controlled, with respect to amounts of intake. This replication of Kalat and Rozin's (1971) findings seriously qualified Revusky's assertion that interference is the single parameter of learning and forgetting in taste-aversion learning. However, it must be noted that interference does preclude learning under certain stimulus conditions.

Looking at the coffee aversion in both experiments, and under both experimental conditions, it may be noticed that when coffee was closer in time to toxicosis, as the last flavor presented, strong aversions were obtained. However, when it preceded three other flavors as the first flavor presented, animals failed to show aversion, compared to controls. Looking at the sucrose aversion in both experiments, and under both conditions, it may be observed that aversions were demonstrated, regardless of whether it was the first or the last flavor presented. Thus, while three interfering flavors produced a strong enough effect to block conditioned aversion to coffee, sucrose appeared to be less sensitive to the interference of three novel flavors. This may suggest that stimuli of different taste quality may be differently influenced by interfering events during the delay between injection and toxicosis. In the light of data which show that different tastes may demonstrate differential associability to toxicosis (Kalat & Rozin, 1970), that taste quality may determine adequacy as conditioned stimulus (Brackbill et al, 1971), and that the natural preference level could be a factor in producing differential aversion (Sutker, 1971), the above suggestion gains further feasibility. Perhaps the quality of interfering flavors is important to consider as well.

Although this study was not designed to determine the qualitative stimulus values in interference, the data form a descriptive picture of a complex relationship

between stimulus quality, amounts of consumption, and order of presentation as effective parameters of interference.

REFERENCES

- Brackbill, R. M., Rosenbush, S. N., & Brookshire, K. H. Acquisition and retention of conditioned taste aversions as a function of the taste quality of the CS. *Learning & Motivation*, 1971, 2, 341-350.
- Bruning, J. L., & Kintz, B. L. *Computational handbook of statistics*. Illinois: Scott, Foresman, 1968. Pp. 112-115.
- Dietz, M. N., & Capretta, P. J. Modification of sugar and sugar-saccharin preference in rats as a function of electric shock to the mouth. *Proceedings of the 75th Annual Convention of the American Psychological Association*, 1967, 161-162.
- Garcia, J., & Ervin, F. R., Gustatory-visceral and telereceptor-cutaneous conditioning-adaptation in internal and external milieus. *Communications in Behavioral Biology*, 1968, 1A, 389-415.
- Garcia, J., Ervin, F. R., & Koelling, R. A. Learning with prolonged delay of reinforcement. *Psychonomic Science*, 1966, 5, 121-122.
- Garcia, J., Kimeldorf, D. J., & Koelling, R. A. Conditioned aversion to saccharin resulting from exposure to gamma radiation. *Science*, 1955, 122, 157-158.
- Garcia, J., & Koelling, R. A. Relation of cue to consequence in avoidance learning. *Psychonomic Science*, 1966, 4, 123-124.
- Kalat, J., & Rozin, P. "Saliency": A factor which can override temporal contiguity in taste-aversion learning. *Journal of Comparative & Physiological Psychology*, 1970, 71, 192-197.
- Kalat, J., & Rozin, P. Role of interference in taste-aversion learning. *Journal of Comparative & Physiological Psychology*, 1971, 77, 53-58.
- Kimble, G. A. *Hilgard and Marquis' conditioning and learning*, (2nd ed.). New York: Appleton-Century-Crofts, 1961.
- Nachman, M. Learned taste and temperature aversion to lithium chloride sickness after temporal delays. *Journal of Comparative & Physiological Psychology*, 1970, 73, 22-30.
- Revusky, S. H. Aversion to sucrose produced by contingent x-irradiation: Temporal and dosage parameters. *Journal of Comparative & Physiological Psychology*, 1968, 65, 17-22.
- Revusky, S. H. The role of interference in associative memory. In W. K. Honig and H. James (Eds.), *Second annual symposium on animal memory at Dalhousie University*. Halifax, Nova Scotia, Canada: Dalhousie University Press, 1971.
- Revusky, S. H., & Bedarf, E. W. Association of illness with prior ingestion of novel foods. *Science*, 1967, 155, 219-220.
- Rozin, P., & Ree, P. Long extension of effective CS-US interval for anesthesia between CS and US. *Journal of Comparative & Physiological Psychology*, 1972, 80, 43-48.
- Siegel, S. *Nonparametric statistics for the behavioral sciences*. New York: McGraw-Hill, 1956. Pp. 116-126.
- Smith, J. C., & Roll, D. L. Trace conditioning with x-rays as an aversive stimulus. *Psychonomic Science*, 1967, 9, 11-12.
- Sutker, L. The effect of initial taste preference on subsequent radiation-induced aversive conditioning of saccharin solution. *Psychonomic Science*, 1971, 25, 1-2.

NOTE

1. The fact that a median of 2.4 ml may be significantly different from a median of 4.2 ml but not from a median of 4.4 ml may be explained in terms of the nature of the U test where the relative rank order of the scores is considered. A look at the means of the QUANTITY, TIME, and CONTROL groups, which are 3.07 ml, 5.04 ml and 4.65 ml, respectively, may help to clarify the observed relationship.

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