Taste Stimuli: Quality Coding Time

Abstract. *Rats conditioned to avoid drinking 300 millimolar NaCl recognized and rejected this solution within 250 to 600 milliseconds of onset of stimulus, a period containing the phasic portion of the peripheral neural response. They generalized to 500 millimolar NaCl but not to 500 millimolar sucrose. Rejection was based on quality identification neurally encoded within this brief period.*

There is considerable uncertainty concerning the time period needed for the quality of a tasted material to be coded in the impulse patterns of the nerves emerging from the tongue and oral cavity. When solutions are continuously flowed over the surface of the tongue for seconds, as is the custom when studying gustatory neural responses in rat and other vertebrates, a characteristic sequence of changes in electrical activity is observed in taste nerves, such as the chorda tympani. After a latency of 25 to 60 msec (*1–4*), a phasic increase in discharge rate occurs which, depending on the kind and concentration of solution, may last from 200 to 400 msec. A prolonged tonic component, lasting several seconds, or minutes, follows (*1, 4, 5*). The magnitudes of these responses have been used for the modeling of quality coding in vertebrate taste nerves (*6–8*). Responses summed over one or more seconds have been used for such analyses. The phasic portions of the response have either been discarded or grouped with the more prolonged tonic components, thus exploring the quality coding capacity of only the tonic component. This exclusive attention to the tonic component is often accompanied by the tacit assumption that taste quality is not or cannot be coded in the phasic discharge of the early reponse period. Observations of behavior in rats suggest that this may be a specious assumption. In our laboratory we have observed that rats tested for gustatory aversion (*9*) take only one lick or a few licks of the conditioning solution, often with a total volume of less than 20 μl, in usually less than 1 second, and then stop drinking. This study deals with this observation quantitatively. The results suggest that the early phasic response period is of great importance in the neural coding of taste quality (*10*).

When rats are trained to consume their daily fluid ration during a single short period each day and during this time the training fluid—distilled water—is presented intermittently for 10-second periods, the animals tend to lick the fluid at a high and relatively uniform rate during each presentation. Long licking bursts for water at a rate of five to seven licks per second are often observed in well-trained animals. The bursts for water usually last from 4 to 10 seconds without a pause. Duration of licks (from start to end of one lick) ranges from 65 to 75 msec for water; from 48 to 68 msec for other fluids (median). For all licking bursts, the longest interval between licks (from the end of one lick to the start of the next lick) for all fluids tested was 98 ± 2 msec (median $\pm$ standard error of the median). Thus, with such subjects, response times to tasted stimuli can be measured with an accuracy of one interval between licks, or approximately 100 msec.

The above restricted fluid consumption schedule was combined with a radiation-induced gustatory avoidance procedure (*9*) in order to determine how long it takes rats to recognize and stop drinking the specific chemical solution which they were conditioned to avoid. Animals were subsequently tested for temporal characteristics of generalization (*9*) to other chemical solutions. Ten rats were adjusted to the schedule of intermittent presentation of fluid for several days, and then were conditioned to avoid drinking a salt solution [300 m*M* NaCl (analytical reagent) made with water distilled in glass] which they would ordinarily consume as freely as the distilled water (*9*). They were offered this NaCl solution as the primary conditioning solution (PCS) (*9*) every 30 seconds for 10-second periods for a total of 40 presentations. Immediately thereafter they were exposed to 200 r whole-body x-irradiation during a 4-minute period [40 r/min, 220 kv (peak), 15 ma]. On the following day, test day 1, each animal was offered 10-second periods of the PCS fluid, alternated with 10-second periods of distilled water, one presentation every 30 seconds, for a total of six presentations of each fluid. Water was presented first. Licks were detected by lickometer circuits (*11*), totaled on counters, and recorded on magnetic tape. Two of the rats were not adequately conditioned according to our previously established criteria

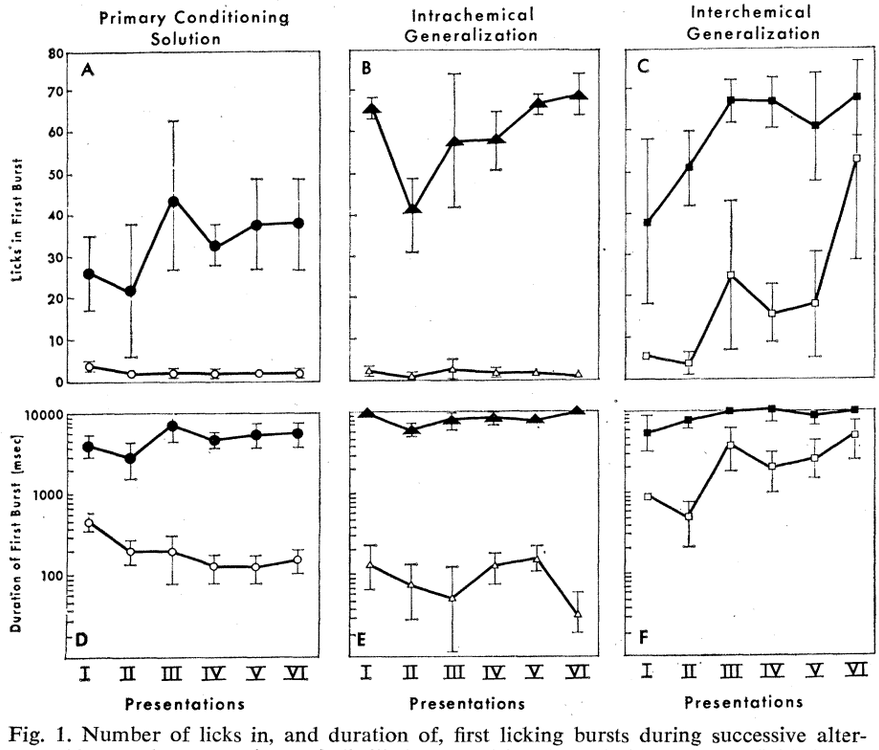

Fig. 1. Number of licks in, and duration of, first licking bursts during successive alternate 10-second presentations of distilled water (closed symbols) and test fluids (open symbols); (A and D) 300 m*M* NaCl; (B and E) 500 m*M* NaCl; (C and F) 500 m*M* sucrose. Plotted values are medians $\pm$ standard error of the median.

(*12*) and were removed from the study. Eight animals strongly rejected the PCS but continued to drink the distilled water (Fig. 1A), consuming only about 6 percent as much 300 m*M* NaCl as water. During the first water presentation they took 46 ± 8 licks (median $\pm$ standard error of the median) (Fig. 2A). Of these, 26 licks occurred during the first licking burst (*13*), which lasted for more than 4 seconds (Fig. 1D). After a 0.5-second pause, the animals resumed drinking. Similar drinking patterns were observed for all six water presentations; the first licking bursts were long and of uniform licking rate. The interval between these licks for water was 82 ± 3 msec.

In contrast to their avid consumption of distilled water, the rats took little of the 300 m*M* NaCl solution. During the first presentation of the PCS fluid, they took 6 ± 1 licks. Four of these occurred in a 500-msec initial burst early in the presentation period (Fig. 1, A and D, and Fig. 2A). After a long pause, 9.3 ± 1.2 seconds, two additional licks were recorded. During successive NaCl presentations, fewer licks were taken and less time was required for the animals to recognize the aversive taste and to reject the fluid. The interval between licks of the PCS was 88 ± 4 msec. By adding this interlick interval time to the duration of the licking burst, an outside estimate of median recognition plus reaction time may be obtained. Thus, it took less than 600 msec for the animals to respond on the first NaCl presentation; there was sufficient information in less than 250 msec of the sixth presentation of the PCS for the animals to cease licking. Quantitatively similar results have been obtained with 500 m*M* DL-alanine as the PCS.

On the first presentation of the PCS, enough information was coded in the early neural discharge from the nerves of the tongue and oral cavity for the animals to distinguish this solution from distilled water. A total of 20 μl of the PCS, applied in four 65-msec pulses, that is, four 5-μl licks (*14*) of that median duration, was sufficient stimulus for this purpose. On subsequent presentations even less volume and time were needed for the animals to make this decision. The results of experiments run the next day (test day 2) indicate that these rats did not nonspecifically detect and reject substances other than water but rather that they made a specific identification of a class of substance (*9*), that is, a quality recognition.

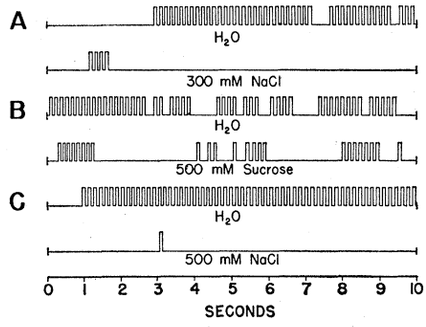

Fig. 2. Licking patterns of rats to initial presentations of 300 m*M* NaCl, 500 m*M* sucrose, or 500 m*M* NaCl, and the preceeding (A) or following (B and C) initial H_2O presentation. One animal with initial presentation of PCS (A) and of interchemical generalization test solution (B). (C) Another animal (also first tested with alternated H_2O and PCS—but record not shown) with initial presentation of intrachemical generalization test solution. Records were traced over optical records of licking patterns reproduced from magnetic tape.

On test day 2, the eight rats were divided into two groups and were tested for temporal characteristics of generalization of their aversion to 300 m*M* NaCl solution with two other solutions. One group was tested for intrachemical generalization to 500 m*M* NaCl and the other for interchemical generalization to 500 m*M* sucrose (analytical reagent, made with water distilled in glass) (*9*). The fluids were offered on the same presentation schedule as on test day 1 with the exception that the test solutions, rather than water, were offered first. The animals tested on the 500 m*M* NaCl solution (intrachemical group) strongly generalized from their aversion to the 300 m*M* solution. On the first presentation they rejected the 500 m*M* NaCl after two licks in a 150-msec licking burst (Fig. 1, B and E); 2 ± 0.4 licks occurred 3.1 seconds later. Since the interval between licks to 500 m*M* NaCl was 98 ± 2 msec, the rats recognized and reacted to the taste stimulus on this and subsequent presentations in less than 250 msec after onset of stimulus, while consuming the distilled water at high lick rates and with short pauses (Fig. 2C).

The animals tested for interchemical generalization reacted to the sucrose solution differently than they did to either water or the 300 m*M* NaCl. On the first presentation they took a total of 7 ± 3 licks, 6 ± 1 of which occurred in a 925-msec initial burst (Fig. 1, C and F). One additional lick was recorded in the middle of the period. During this first association with the sucrose solution they licked seven times longer before stopping than the rats given the 500 m*M* NaCl (Fig. 2B). We believe that this represents the beginning of not only a *not water* but also of a *not PCS* categorization. During the second presentation of 500 m*M* sucrose the rats took a total of 24 ± 8 licks in two bursts. By the third presentation, they drank for a prolonged initial burst, 4.0 seconds, indicating, behaviorally, that the sucrose solution, while differing from distilled water, was not in the same quality category as the NaCl solutions.

The licking patterns illustrate that these rats grouped the distilled water, NaCl, and sucrose solutions into separate classes. While this was most apparent from licks summed over the total presentation series, it was already evident during the first presentation of each solution. Throughout both test days the water was freely consumed, with 52 licks per period on day 1 and 65 on day 2. The PCS was rapidly recognized and rejected during 600 msec of the first presentation. Even less stimulus input time was required to convey this information on subsequent offerings and this reduced time was adequate to inform the animals of the similarity with the 500 m*M* NaCl solution on day 2. In contrast, rats primed to reject the PCS in less than 250 msec licked the novel sucrose solution for approximately 925 msec on the first presentation, and on the remaining presentations increased consumption of this fluid. All the neural cues enabling the animals to distinguish these different fluids apparently occurred during the early response phase of the neural discharge, within less than 1 second of onset of stimulus. Since central analysis, decision-making, and motor activity take up a significant portion of the total time needed to stop the licking behavior, and may continue long after the removal of a stimulus source (*15*), it is plausible that the quality of the taste was coded in the peripheral afferent input in a much shorter period after onset of stimulus.

The initial quality coding time observed for rats in this experiment (600 msec) corresponds to reports of human

taste quality reaction time (*16*). In addition, when humans are presented NaCl or sucrose solutions for 700 to 800 msec, they receive enough information to quantitatively estimate concentration (*17*).

Most neurophysiological models of taste quality categorization and of gustatory receptor kinetics (*2*, *7*) have not systematically utilized the activity from the early phasic response period. It is well known in other sensory systems that the transient or phasic portions of activity are most important, and incorporation of these behaviorally relevant temporal properties of gustatory response into such models may produce an improved understanding of taste and other sensory systems.

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10. The observed rapid cessation of licking is thought to depend on gustatory afferent input for several reasons. First, complete absence of licking during a presentation occurred less than 8 percent of the time, and was found equally often for water and the NaCl solutions. Thus, contact of the tongue with the liquid was necessary for further licking to be withheld. Second, duration of an initial licking burst to an aversive solution never exceeded 800 msec, but the latency of a vertebrate olfactory primary neuron response is 1 to 2.8 seconds. Thus, olfactory afferent input would not occur until several hundred milliseconds, or more, after the licking burst had ended [R. J. O'Connell and M. M. Mozell, *J. Neurophysiol.* **32**, 51 (1969)]. Third, the general vapor phase environment of the drinking tubes was made relatively homogeneous by locating the tubes close to each other.
11. Licking was done by extending the snout through a single opening 1 cm in diameter. The two stainless steel drinking tubes were alternately positioned 2 mm outside this opening by an automatic mechanism. The tubes were fixed so that the nonpresented tube was 2.2 cm above or below the presented tube at all times. Between presentations, access was prevented by an automatic shutter. The lickometer circuits were Grason-Stadler E4690A-Z1 Rev. O, which passed less than 1 μa through the animal.
12. Each animal must consume at least 80 percent as much water at each presentation as it consumed on the day prior to conditioning. Its PCS consumption must be less than 15 percent of water consumption for each of the six consecutive water-PCS presentation pairs. Radiation-induced gustatory avoidance of NaCl (140 m*M*) has previously been reported [N. W. Perry, Jr., *Radiat. Res.* **20**, 471 (1963)].
13. The first licking burst of each period was considered ended when an interlick interval of 200 msec, or of twice the median interlick interval of the first five licks with intervals less than 200 msec, whichever was greater, occurred. A "pause" in 10-minute licking periods, in which the interlick interval was 150 ± 15 msec (mean ± standard deviation), has been defined as 200 msec [J. D. Corbit and E. S. Luschel, *J. Comp. Physiol. Psychol.* **69**, 119 (1969)].
14. The volume of individual licks was measured by dividing the total number of licks into the total volume consumed. A value of 5 ml had been previously reported [E. Stellar and J. H. Hill, *J. Comp. Physiol. Psychol.* **45**, 96 (1952)].
15. The latency for rat chorda tympani single unit responses to gustatory stimulation with sucrose has been reported to be as long as 300 msec (*3*) or as short as 90 msec [M. S. Nejad, "Factors involved in the mechanism of stimulation of gustatory receptors and bare nerve endings of the tongue of the rat," Ph.D. thesis, Florida State University (1961)]. Experiments using a digitalized summator with 10 msec integrating intervals to discretely analyze chorda tympani population responses, and a calibrated reflection phototransistor to determine liquid contact with the tongue, indicate a latency of 60 msec; J. R. Faull and B. P. Halpern, in preparation. For digitalized summator, see A. D. Brush and B. P. Halpern, *Physiol. Behav.* **5**, 743 (1970); for calibrated phototransistor, see (*4*). Visual stimulus presentations of 50 msec are still being processed 100 msec or more later. See U. Neisser, *Cognitive Psychology* (Appleton-Century-Crofts, New York, 1967), pp. 15–27.
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Melatonin: Effect on Punished and Nonpunished Operant Behavior of the Pigeon

Abstract. *Intramuscular injections of melatonin had a dose-dependent, rate-increasing effect on responding maintained by a fixed-interval schedule of positive reinforcement. When a punishment contingency was added to the fixed-interval schedule, the overall rates were not increased although responding was increased during the initial part of each interval.*

Melatonin, a major constituent of the pineal gland (*1*), has effects on many endocrine systems (*2*, *3*). In addition, systemic or intracerebral administration of melatonin to cats or systemic administration to young chickens produces sleep and corresponding changes in the electroencephalogram (*4*). A slight decrease in motor activity occurs after administration of low doses to rats (*5*). In a study of the central metabolic effects of melatonin (*6*), both increased and decreased levels of serotonin were found after intraperitoneal injection. The direction of the change was dependent upon the brain region sampled. This finding and the experimental work implicating serotonin in the mechanism underlying sleep production (*7*) have led to the suggestion that serotonin derivatives found in the pineal gland readily gain access to the central nervous system where they modify the function of serotonin-containing neurons (*3*).

In the following experiments I have studied the effect of melatonin on punished and nonpunished key-pecking behavior of the pigeon. It has thus been possible to determine the behavioral effects of melatonin in situations in which a wide variety of compounds has been studied (*8*). The present study also makes possible a comparison of the behavioral effects of melatonin with those of agents presumed to alter the function of central neurons containing serotonin, since punished and nonpunished schedules of reinforcement have been previously utilized to study the behavioral effects of serotonin agonist and antagonist drugs (*9*).

Eight adult, male, White Carneaux pigeons were housed in individual cages and maintained at 80 percent of their free-feeding weights. All birds had been previously trained and used in drug experiments. Four birds were trained on a multiple fixed-interval 5-minute fixed-ratio 30-response schedule (multiple FI 5 FR 30) and the other birds were trained on a concurrent fixed-interval 5-minute (food) fixed-ratio 30 (shock) schedule (concurrent FI 5 FR 30).

The experimental chamber was similar to that described by Ferster and Skinner (*10*). A translucent key, 2 cm in diameter, was mounted in the partition wall of the chamber facing the animal compartment. The key could be transilluminated with lights of different colors. The minimum force required to operate the key was about 15 g. A rectangular opening below the response