Responses to Binary Taste Mixtures in the Nucleus of the Solitary Tract: Neural Coding With Firing Rate

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Chen J-Y, Di Lorenzo PM. Responses to binary taste mixtures in the nucleus of the solitary tract: neural coding with firing rate. J Neurophysiol 99: 2144–2157, 2008. First published February 20, 2008; doi:10.1152/jn.01020.2007. The contribution of gustation to the perception of food requires an understanding of how neurons represent mixtures of taste qualities. In the periphery, separate groups of fibers, labeled by the stimulus that evokes the best (largest) response, appear to respond to each component of a mixture. In the brain, identification of analogous groups of neurons is hampered by trial-to-trial variability in response magnitude. In addition, convergence of different fiber types onto central neurons may complicate the classification scheme.

To investigate these issues, electrophysiological responses to four tastants: sucrose, NaCl, HCl, and quinine, and their binary mixtures were recorded from 56 cells in the nucleus of the solitary tract (NTS, the 1st synapse in the central gustatory pathway) of the anesthetized rat. For 36 of these cells, all 10 stimuli were repeated at least five times (range: 5–23; median = 10). Results showed that 39% of these cells changed their best stimulus across stimulus repetitions, suggesting that response magnitude (firing rate) on any given trial produces an ambiguous message. Averaged across replicate trials, mixture responses most often approximated the response to the more effective component of the mixture. Cells that responded best to a taste mixture rather than any single-component tastant were identified. These cells were more broadly tuned than were cells that responded best to single-component stimuli and showed evidence of convergence from more than one best stimulus fiber type. Functionally, mixture-best cells may amplify the neural signal produced by unique configurations of basic taste qualities.

INTRODUCTION

To appreciate the contribution of gustation to the perception of food, it is important to understand how taste is encoded by the nervous system. Although it has been argued that the “taste world” is organized around a small number of similar-tasting groups, called the “basic” taste qualities (sweet, sour, salty, bitter, and perhaps umami), most natural foodstuffs do not easily fit into a single category; instead, most foods are mixtures of taste qualities. So a comprehensive theory of taste coding must include an account of the neural representation of these complex tastants.

Thus far data from studies of the electrophysiological responses to taste mixtures in rodents are generally consistent with the idea that taste mixtures excite separate subsets of cells, each sensitive to one of the components of the mixture (Travers and Smith 1984). Conversely, cells that respond most vigorously to a particular taste quality (defined as its “best” stimulus) show distinctive and predictable responses to the various binary taste mixtures (Frank 1989; Hyman and Frank 1980b; Travers and Smith 1984; Vogt and Smith 1993a,b). These data support the widely held contention that taste mixtures are essentially combinations of identifiable and separable basic taste qualities that do not evoke any taste sensations other than those of the components. Behavioral data demonstrating that conditioned aversions to taste mixtures will generalize almost exclusively to the components of the mixtures are consistent with this idea (Formaker and Frank 1996; Nowlis and Frank 1981; Smith and Theodore 1984).

The idea that separate subsets of neurons exist that can be used to detect the components of a taste mixture may be intuitively satisfying but leaves several issues unresolved. First, there is the question of how to identify these subsets. While the best stimulus classification scheme has its origins in studies of afferent nerves, many would argue that it offers a reasonable way to organize taste sensitivities in central gustatory structures as well (reviewed in Spector and Travers 2005). For example, the same organization of best-stimulus fiber types that exists in peripheral nerves is also apparent in the nucleus of the solitary tract (NTS, the 1st central relay in the gustatory pathway) (Nakamura and Norgren 1991, 1993), even though NTS cells are more broadly tuned (they respond to >1 taste quality) than peripheral nerve fibers (Doetsch and Erickson 1970). In particular, when NTS cells are classified either by their best stimulus or by hierarchical cluster analyses, separate classes of neurons that respond best to sugars, sodium salts, alkaloids, and electrolytes emerge. Furthermore, when intensity-response functions have been examined, the slope of these functions for a neuron’s best stimulus is steeper than for nonbest stimuli in a given best-stimulus class (Nakamura and Norgren 1991; Nishijo and Norgren 1990). On the other hand, statistical analysis of response patterns in the NTS using receiver-operator curves have demonstrated that cell types alone cannot convey enough information to identify a tastant unambiguously (Lemon and Smith 2006).

A second issue concerning coding of taste mixtures is the question of how information from separate “channels” of information (most likely stimulus-best fibers types) are integrated when a complex taste is presented. There are at least two potential solutions to this problem. First, based on the notion that a given stimulus-best cell is charged with the task of encoding stimuli of a single taste quality, e.g., activity in sucrose-best cells signals sweetness, but not saltiness, etc., it can be hypothesized that a binary taste mixture will excite a given neuron to the extent that the stimulus contains its best
Real-time records of neural activity were saved for off-line analysis. In that case, the response to a binary taste mixture would be equivalent to the response to the more effective components (MECs) of the mixture. In a second way of thinking, afferent signals arising from different taste qualities might converge onto the same cell. In that case, one might expect the response to a binary mixture to be “additive” such that it will be equal to the sum of the responses to each of the components, and cell types might be defined by the best single or best combinations of taste qualities that excite them.

In the present study, we describe electrophysiological responses to NaCl, sucrose, HCl, and quinine as well as their undiluted binary mixtures in single cells in the NTS of the anesthetized rat. This is the first report on NTS responses to mixtures thus far in the literature. Our analyses addressed three issues concerning the neural representation of mixtures in the brain stem. First, we assessed the reliability of classification of NTS cells according to their best stimulus. Second, we compared mixture responses to responses to the MEC and to the sum of responses to each of the components of the mixture to examine how inputs from single tastants are integrated in mixture responses in NTS cells. Finally, in the context of stimulus repetition, we compared the reliability of the classification of individual cells into groups with the reliability of across neuron patterns to each stimulus.

METHODS

Subjects

Thirty-five adult male Sprague-Dawley rats (300–450 g) were subjects in this experiment. Rats were pair housed in plastic cages and maintained on a 1-h light-dark schedule with lights on at 7:00 a.m. Food and water were available ad libitum. All procedures were in accord with the National Institutes of Health animal welfare guide and were approved by the Institutional Animal Care and Use Committee of Binghamton University.

Surgery

Rats were anesthetized with urethan (1.5 g/kg ip administered in 2 equal doses spaced 30 min apart) and prepared surgically for electrophysiological recording in the NTS. Animals were tracheotomized and mounted in a stereotaxic instrument so that the head was positioned with the tooth bar 5 mm below the interaural line. This position allowed access to the NTS without disruption of any sinuses or major blood vessels. The occipital bone was removed and uvular and nodular portions of the cerebellum were aspirated gently to expose the dorsal surface of the caudal medulla. A nontraumatic head holder was cemented to stainless steel screws embedded in the skull using dental acrylic. Core temperature was monitored with a rectal thermometer linked to a heating pad and maintained at 37°C.

Recording

Taste-responsive cells in the NTS were isolated with tungsten microelectrodes (18–20 MΩ at 1 kHz; FHC, Bowdoinham, ME). Signals were fed to an amplifier (Model P511, Grass Technologies, West Warwick, RI) and digitized as described in the following text. Real-time records of neural activity were saved for off-line analysis. The electrode was positioned above the NTS and lowered slowly into the medulla. Previous work in our lab has demonstrated that the taste-responsive portion of the NTS is located ~2.7 mm rostral and 1.8 mm lateral to the obex and between 0.7 and 1.4 mm below the surface of the brain stem. As the electrode was advanced into the medulla, the presence of taste responses was tested by bathing the tongue with 0.1 M NaCl (5 s) followed by a rinse of distilled water (20 s). To prevent a sampling bias in the cells chosen for analysis, once the electrode detected a response to NaCl in the background, every isolated cell was tested with all four of the basic taste stimuli (see following text).

Taste stimuli and stimulus delivery

Tastants representing the four basic taste qualities and their binary mixtures were presented as stimuli. Single-component taste stimuli consisted of NaCl (0.1 M; N), sucrose (0.5 M; S), quinine HCl (0.01 M; Q), HCl (0.01 M; H). The six binary mixtures (NH, NS, NQ, HS, HQ, SQ) each consisted of components whose final concentration in the mixture equaled that of the single-component taste stimuli. These concentrations were chosen because they produce half-maximal responses in the chorda tympani nerve (CT; a branch of the facial nerve innervating taste buds on the rostral 2/3 of the tongue) of the rat (Ganchrow and Erickson 1970; Ogawa et al. 1974). All stimuli were made with reagent grade chemicals, dissolved in distilled water and presented at room temperature.

Taste stimuli were bathed over the tongue through a specially designed stimulus delivery system described previously (Hallock and Di Lorenzo 2006). Briefly, tastants were delivered through six separate perforated stainless steel tubes. Stimulus delivery was controlled by a computer that operated solenoid valves positioned between pressurized stimulus reservoirs and the stainless steel tubes. The flow rate was 5 ml/s.

Testing

Once a taste-responsive cell was isolated, each of 10 tastants, i.e., four single-component taste stimuli (N, S, Q, and H) and six binary mixtures (NH, NS, NQ, HS, HQ, SQ), were presented in individual trials. Each trial consisted of a 10-s baseline (spontaneous activity), 10-s distilled water prerinse, 5-s presentation of the tastant, 5-s pause, and 20-s distilled water rinse. The interstimulus interval was 2 min. Two or three blocks of the four single-component tastants were alternated with two or three blocks of the six binary mixtures in a pseudorandom fashion for as long as the cell remained well isolated. Stimulus delivery tubes were flushed well with distilled water when the stimulus to be delivered was changed.

Data analysis

Identification of single cells was accomplished using specialized software ( Spike2; CED, Cambridge, UK; sampling rate = 25 kHz). Waveforms arising from single cells were recognized using a waveform template-matching algorithm. Signal-to-noise ratios were ≥3:1. Action potentials were stamped with the time of occurrence (resolution = 1 ms) relative to the beginning of each stimulus trial.

Response magnitude was measured as the rate of firing in spikes per second (SPS) in the 5 s of stimulus presentation minus the firing rate in the final 5 s of the water prerinse. A change in the average firing rate over the first 5 s of stimulus presentation that differed from the average firing rate during the water prerinse (last 5 s) by ≥2.54 SD was defined as a significant response.

NTS cells were classified initially according to their “best stimulus,” i.e., the single-component tastant that evoked the most vigorous response regardless of the amount with which the response to the best stimulus exceeded the response to the next-best stimulus.

In cases where a binary mixture was the best stimulus, statistical criteria were used to define mixture-best cells. For 36 cells, the availability of many responses to each stimulus enabled the assessment of statistical reliability. For these cells, an ANOVA of all responses was conducted with the significance of pairwise comparisons determined by a Bonferroni post hoc test. A mixture-best cell was defined as a cell with a response to a taste mixture that was...

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significantly greater ($P < 0.05$) than the response to either component presented individually. For the remaining 20 cells, a cell with a response to a mixture that was $\geq 5$ SPS (for cells with mean responses of $\geq 5$ SPS) or 100% (for cells with mean responses $< 5$ SPS) greater than the response to the MEC was considered to be mixture-best.

To examine the breadth of tuning of taste-responsive NTS units, an Uncertainty measure (Smith and Travers 1979) was calculated for each cell as follows

$$H = -K \left( \sum_{i=1}^{n} P_i \log P_i \right)$$

where, $P_i$ represents the number of spikes elicited by each stimulus expressed as a proportion of the total number of spikes elicited by $n$ stimuli, and $K$ is a scaling constant. For four stimuli, $K = 1.661$, which results in $H$ ranging from a minimum of 0 (cell responds to only 1 stimulus) to a maximum of 1.0 (cell responds equally well to all stimuli). Mean Uncertainty values are expressed as the means $\pm$ SE.

To examine the organization of response profiles from individual cells, hierarchical cluster analyses were used. This analysis begins with a matrix of similarities among the responses across stimuli for each cell. Pearson product-moment correlations were used to measure similarity in the present study. The analysis then proceeds in a stepwise fashion: initially, the two most similar response profiles are joined into a cluster. Then the next two most similar response profiles, or the response profile most similar to that initial cluster and the initial cluster, are joined. As this process continues, new clusters are formed and/or new members are added to existing clusters until all elements are joined into a single cluster. The output of this analysis identifies those cells that have response profiles most similar to each other, and therefore might be considered part of a group or type. The SYSTAT program on a PC-compatible computer was used for the cluster analyses. The Ward minimum variance method was used as a cluster algorithm.

To examine the organization of across neuron patterns (ANPs) of response evoked by each stimulus, multidimensional scaling (MDS) analysis was used. This analysis places taste stimuli within a hypothetical “taste space” based on the degree of similarity (measured as the Pearson product-moment correlation) of their evoked response magnitudes across neurons. Stimuli that evoke a similar ANP of response are placed close to each other and those with dissimilar magnitudes across neurons. Stimuli that evoke a similar ANP of response are placed relatively far apart. The SYSTAT program was used for this analysis.

RESULTS

Electrophysiological responses to taste stimuli were recorded from 56 single NTS cells. For 36 of these cells, all 10 stimuli were presented at least five times (range: 5–23 presentations; median = 10). For the remaining 20 cells, responses to at least one presentation of all 10 taste stimuli were recorded, but it was not possible to repeat all stimuli more than a few times (range: 1–8 presentations; median = 2). Average spontaneous rate was 2.15 ± 2.46 SE SPS. The order of effectiveness, assessed as the average firing rate of response across all cells, among all taste stimuli was NH > N > NS > NQ > HS > H > SQ > HQ > Q > S. Among the four single taste stimuli, there were 55 cells (98%) that responded to N, 53 cells (95%) to H, 46 cells (82%) to Q, and 34 cells (61%) to S. All taste mixtures evoked responses in the great majority of cells (range: 51 cells for HQ and SQ to 56 cells for NS and NQ).

Taste-responsive cells were generally broadly tuned across tastants (mean Uncertainty = 0.71 ± 0.02). When only the four single tastants were considered, there were 28 cells (50%) that responded to all four tastants, 23 cells (41%) that responded to three tastants, three cells (5%) that responded to two tastants, and only two cells (4%) that responded to a single tant. For taste mixtures, 48 cells (86%) responded to all six mixtures, three cells (5%) to five mixtures, two cells (4%) to four mixtures, and three cells (5%) to three mixtures.

Figure 1 shows the mean firing rate evoked by all 10 tastants across all cells. Mean responses differed significantly across tastants [F(9,55) = 47.0, $P < 0.01$]. Planned comparisons of single tastants with binary mixtures containing those tastants showed that there were no significant differences between N and mixtures containing N (Bonferroni post hoc test, $P > 0.05$). Also, responses to H were not different from those to HS nor were the responses to S or Q significantly different from those to SQ ($P > 0.05$). Interestingly, the response to HQ was significantly smaller than the response to H even though H was the MEC of this mixture (that is, the component that produced the larger response when presented alone).

The repetition of taste stimulus presentations in 36 cells permitted the analysis of variability in response magnitudes. When only the four single tastants were considered, most taste-responsive cells showed a consistent best stimulus across blocks of trials (22 of 36; 61%). The remainder, 14 cells (of 36; 39%), changed their best stimulus in some proportion of blocks of trials (mean = 26 ± 3% blocks of trials; range of 14–50%; median = 25%). Of these 14 cells, 10 (71%) were classified as mixture best when responses to all 10 stimuli were considered. Of the 22 cells with reliable single-component best stimuli, 4 (18%) were classified as mixture best when responses to all 10 stimuli were considered.

Because a given stimulus produced a wide range of response magnitudes across cells, a measure of variability that was scaled according to the average response magnitude was applied to all responses where replicate trials were available. This measure was the coefficient of variation (CV), calculated as the ratio of the SD to the mean response. The mean CV across all cells was 0.32 ± 0.03. N and mixtures containing N were the least variable across trials (all mean CVs were 0.18 ± 0.03) while S (CV = 0.44 ± 0.01), Q (CV = 0.43 ± 0.01), and the mixture of SQ (CV = 0.44 ± 0.01) were the most variable. A repeated-measures ANOVA of all CVs in cells that had significant responses to all tastants and taste mixtures ($n = 27$)
showed a significant main effect of stimulus \( F(9,234) = 14.00, P < 0.001 \); Bonferroni post hoc tests showed that CVs for \( S = Q > N = H \) \( P < 0.01 \). Stimulus-related differences in variability were most likely related to response magnitude because there was a statistically reliable correlation between response magnitude and the CV \( r = -0.47 \): the correlations between response magnitude and CV were significant for all stimuli \( P < 0.05 \) and ranged between \(-0.40 \) for N and \(-0.61 \) for H.

**Responses to taste mixtures compared with responses to the components—how do the components combine?**

Examples of the various types of effects of taste mixtures from three separate cells are shown in Fig. 2. In Fig. 2A, the addition of H to N severely attenuated the response to N when N and H were mixed. This contrasts with the response to NH, shown in Fig. 2B, which was equal to the response to N, the MEC in this case. Figure 2C illustrates an example of mixture enhancement where the response to NQ was greater than the sum of the responses to N or Q presented alone.

If a cell encodes information about both components of a mixture, the response to the mixture should be different from the response to either component presented alone. Because all responses to single tastants in the present study were excitatory, responses to cells that receive information about two tastants might plausibly be close to the sum of the responses to the components, assuming that stimulus interactions, either in the periphery or in the brain, were of minimal impact. In fact, there are well-known stimulus interactions as evidenced in peripheral nerve responses to binary mixtures, at least in hamster (Frank 1989; Hyman and Frank 1980a,b). To assess the extent to which such peripheral interactions are carried forward to cells in the CNS, we conducted an analysis of NTS responses aimed at assessing how close responses to taste mixtures were to the sum of the responses to the components. Specifically, we plotted the sum of the responses to the components of a mixture against the actual response to the mixture. For cells where more than one response to any stimulus was recorded, we used the average response across trials. Linear regression was then applied to these responses. Results are shown in Fig. 3, A and B. Across all cells, repeated-measures ANOVA of all responses to mixtures and the sum of the responses to the components showed a significant effect of stimulus \( F(11,605) = 50.21, P < 0.01 \). Pairwise comparisons using the Bonferroni post hoc test showed that responses to NH, NQ, and HQ were significantly less than the sum of the responses to the components \( P < 0.01 \); responses to NS, HS, and SQ were not significantly different from the sum of the responses to the components \( P > 0.05 \).

Another approach to the analysis of responses to taste mixtures is based on the assumption that if a given cell encodes information about only one stimulus of a binary mixture, then the response to that mixture should show no evidence of the addition of the second stimulus; that is, no information about the second stimulus would be encoded. Therefore the response to the mixture should equal the response to the MEC. Accordingly, we plotted the response to the MEC against the actual response for each mixture. Linear regression was then applied to these responses. Results are shown in Fig. 3, C and D. A repeated-measures ANOVA of all responses to mixtures and the MECs for all mixtures revealed a significant effect of stimulus \( F(11,605) = 38.57, P < 0.01 \). Responses to HQ were significantly less that the response to the MEC \( P < 0.02 \), but responses to all other mixtures (NH, NS, NQ, HS, and SQ) did not differ significantly from their MEC \( P > 0.05 \).

For individual cells, responses to taste mixtures were compared with responses to the MEC (Table 1A) or to the sum of the response to the components (Table 1B) of the mixtures. For the 36 cells where sufficient replications were available for tests of statistical reliability, mixture effects were assessed using an ANOVA with the significance of pairwise comparisons determined by a Bonferroni post hoc test. For the remaining 20 cells, changes of \( \geq 5 \) SPS (for cells with mean responses \( \geq 5 \) SPS) or 100\% (for cells with mean responses \(< 5 \) SPS) were considered to be significant.

Comparisons of responses to mixtures with responses to the MEC or to the sum of the responses to the components of the mixture showed that inputs generated by single-component tastants combined in a variety of ways in the NTS even within the same cell. The great majority of cells (52 of 56, 93\%) showed responses to one or more taste mixtures that were significantly greater than the sum of the responses to the MEC or to the sum of the responses to the components of the mixture. For individual cells, responses to taste mixtures compared with responses to the MEC or to the sum of the responses to the components of the mixture were significantly different from the sum of the responses to the components of the mixture. For the 36 cells where sufficient replications were available for tests of statistical reliability, mixture effects were assessed using an ANOVA with the significance of pairwise comparisons determined by a Bonferroni post hoc test. For the remaining 20 cells, changes of \( \geq 5 \) SPS (for cells with mean responses \( \geq 5 \) SPS) or 100\% (for cells with mean responses \(< 5 \) SPS) were considered to be significant.

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either significantly larger or smaller than the response to the MEC and a similar majority (51 of 56, 91%) showed responses to at least one taste mixture that differed from the sum of the responses to the components. Although 201 of 336 mixture responses (60%) were not different from the response to the MEC, 50 mixture responses (15%) were significantly larger, and 85 mixture responses, 25%, were significantly smaller than the response to the MEC. When responses to taste mixtures were compared with the sum of the responses to the components, 188 responses (56%) were not different; the great majority (167 of 172 responses, 97%) of those that were different were significantly less.

To summarize the analyses of comparisons of responses to taste mixtures with the MEC or to the sum of the single-component responses, most responses to taste mixtures were best predicted by the response to the MEC. The single exception to this statement is the response to HQ, which was smaller than both the response to the MEC and the sum of the responses to H and Q. Although responses to NS, HS and SQ were not significantly different from the response to the single-component stimuli, this result is likely due to the relatively small response magnitude of sucrose, such that the sum of the responses to the components is nearly equal to the response to the MEC.

An important observation was a paradoxical effect of some tastants when presented as part of a binary mixture. For example, in seven cells (13%), there was a response to a single tastant, but no response to a mixture containing that stimulant. These included four cells where there was a significant response to H but no response to HQ; two of these cells also responded to Q. Three cells showed a response to Q but no response to SQ; one of these also responded to S. Conversely, there were 20 cells (36%) that showed no response to S (n = 11; 20%), Q (n = 8; 14%), or H (n = 1; 2%).

### TABLE 1. Responses to taste mixtures

<table>
<thead>
<tr>
<th></th>
<th>NH</th>
<th>NS</th>
<th>NQ</th>
<th>HS</th>
<th>HQ</th>
<th>SQ</th>
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<tbody>
<tr>
<td>A.</td>
<td></td>
<td></td>
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<tr>
<td>Number of cells</td>
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</tr>
<tr>
<td>Enhanced</td>
<td>15 (27)</td>
<td>5 (9)</td>
<td>3 (5)</td>
<td>15 (27)</td>
<td>4 (7)</td>
<td>8 (14)</td>
</tr>
<tr>
<td>Suppressed</td>
<td>12 (21)</td>
<td>13 (23)</td>
<td>15 (27)</td>
<td>3 (5)</td>
<td>29 (52)</td>
<td>13 (23)</td>
</tr>
<tr>
<td>Mean spikes per second (sps)</td>
<td></td>
<td></td>
<td></td>
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</tr>
<tr>
<td>Enhanced</td>
<td>11.8 ± 1.7</td>
<td>8.2 ± 2.1</td>
<td>9.4 ± 5.5</td>
<td>9.8 ± 1.5</td>
<td>3.9 ± 1.2</td>
<td>5.8 ± 1.6</td>
</tr>
<tr>
<td>Suppressed</td>
<td>10.9 ± 2.5</td>
<td>10.5 ± 1.7</td>
<td>9.0 ± 1.4</td>
<td>7.4 ± 2.3</td>
<td>11.4 ± 1.1</td>
<td>7.8 ± 1.4</td>
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<tr>
<td>B.</td>
<td></td>
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<tr>
<td>Number of cells</td>
<td></td>
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</tr>
<tr>
<td>Enhanced</td>
<td>1 (2)</td>
<td>0</td>
<td>1 (2)</td>
<td>2 (4)</td>
<td>1 (2)</td>
<td>0</td>
</tr>
<tr>
<td>Suppressed</td>
<td>39 (70)</td>
<td>24 (43)</td>
<td>33 (59)</td>
<td>14 (25)</td>
<td>40 (71)</td>
<td>17 (30)</td>
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<tr>
<td>Mean sps</td>
<td></td>
<td></td>
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<tr>
<td>Enhanced</td>
<td>16.3</td>
<td>0</td>
<td>3.7</td>
<td>14</td>
<td>2.9</td>
<td>0</td>
</tr>
<tr>
<td>Suppressed</td>
<td>16.4 ± 1.7</td>
<td>12.4 ± 1.3</td>
<td>12.2 ± 1.3</td>
<td>7.2 ± 0.7</td>
<td>15.9 ± 1.6</td>
<td>10.4 ± 1.4</td>
</tr>
</tbody>
</table>

Values in parentheses are percentages; sps values are means ± SE. NH, NaCl plus HCl; NS, NaCl plus sucrose; NQ, NaCl plus quinine; HS, HCl plus sucrose; HQ, HCl plus quinine; SQ, sucrose plus quinine.
2%) but showed either enhanced or suppressed responses when these ineffective stimuli were presented with other tastants in a mixture.

Taste mixtures, cell types, and mixture-best cells—are some cells specialized for signaling mixtures?

To investigate the effects of mixtures on cell types, we first classified cells with respect to their responses to the four single tastants and then considered responses to taste mixtures. When only the single tastants were considered, results showed that there were 41 N-best, 8 H-best, 2 Q-best, and 5 S-best cells. However, when we considered responses to taste mixtures as well as responses to single tastants, many cells responded best to one or more binary mixtures than they did to any tantant presented alone. Analyses of response magnitudes across trials showed that there were 21 cells (38%) that were mixture-best: 11 NH best, 5 HS best, 3 NQ best, and 2 NS best. Of the remaining cells, 28 were N best, 3 were H best, and 4 were S best; there were no Q-best cells.

Mixture-best cells could be distinguished from other cell types by their response profiles and breadth of tuning. For example, responses to the best mixture in mixture-best cells were on average $10.3 \pm 1.5$ SPS larger than the responses to the MEC of that mixture. This represents a $49 \pm 6\%$ increase in response magnitude above the response to the MEC (median $= 46\%$; range: 15–110\%). Most responses to the best taste mixture in mixture-best cells approximated the sum of the responses to the two components of their best mixture presented alone. However, in three mixture-best cells (of 21; 14\%), responses to their best taste mixture were significantly greater ($P < 0.01$; mean $= 14.8 \pm 3.5$ SPS; $66 \pm 15\%$) than the sum of the responses to the components presented separately. There were 2 HS- and 1 NH-best cells that showed this effect. Mixture-best cells were also significantly more broadly tuned than cells that responded best to a single tantant: the average Uncertainty for mixture-best cells was $0.80 \pm 0.03$, average Uncertainty for cells that responded best to a single-component stimulus was $0.68 \pm 0.03$, Student’s $t$-test, $P < 0.01$.

Figures 4 and 5 illustrate the relative responses to taste stimuli in mixture-best cells. In Fig. 4, responses from two NH-best cells (cells 9 and 11) and one HS-best cell (cell 21) are shown across several trials. It is evident from this figure that in all three cells response to their best mixture was significantly and consistently larger than the responses to any other tantants. Figure 5 shows raster plots of responses to all taste stimuli presented nine times in an NH-best cell (cell 7). It can be seen that the responses of this cell to NH were more vigorous and more sustained than the responses to either N or H; the same was true for responses to NS and HS with respect to their component tantants. Interestingly, responses to SQ appeared to be a combination of the responses to each component because Q evoked a short-lived response and S evoked a long latency response in this cell.

The best stimulus classification of an NTS cell was a good predictor of its responses to taste mixtures. These results, presented in Table 2 and in Fig. 6, describe functional differences between single-component-best and mixture-best cells. Comparisons among N-, H-, and NH-best cells showed that suppression of the responses to N by the addition of Q occurred in N-best (12 of 28) and H-best (1 of 3) cells but not in any NH-best cell. Further, suppression of N responses by the addition of H was observed exclusively in N-best cells. The addition of Q to H produced suppression in nearly every NH- and H-best cells but in only about half of the N-best cells. When average response magnitudes across cells were examined, NH responses were greater than N responses ($P < 0.004$) only in NH-best cells; HQ responses were significantly less than H responses ($P < 0.001$) only in H-best cells. All other comparisons of mean responses rates for taste mixtures were not significantly different from the mean responses rates to the MEC of the mixture in these three cell groups. Not surprisingly, responses to taste mixtures in HS-best cells showed a FIG. 4. Response magnitudes (mean spikes over 5 s of response) across trials for 3 mixture-best cells.

FIG. 5. Raster plots of 9 repetitions of all taste stimuli for cell 7.
significantly larger HS response compared with the S response ($P < 0.05$).

To explore the organization of groups of cells with response profiles that (relative response rates across taste stimuli) were similar, we used hierarchical cluster analysis. Results are presented as a dendrogram (Fig. 7) in which the distances between cells or clusters of cells are inversely proportional to the correlations. Cells were labeled according to their best stimulus; the number after the best stimulus label indicates the rank of the response to the best stimulus compared with other cells in the group. Four groups of cells, one associated with each of the four basic taste qualities, were suggested. Within the N group there were four subsets, two of which were narrowly tuned to NaCl (N1 and N2) and two of which were more broadly responsive to both NaCl and HCl (NH1 and NH2). While cells in both N groups responded well to mixtures containing NaCl as a component, both NH groups responded well to both NaCl and HCl. Cells in the NH2 group evidenced some sensitivity to sucrose in their response to SQ. Cells in the H group also responded well to both NaCl and HCl but showed special sensitivity to mixtures containing H.

Results of the hierarchical cluster analysis were in general agreement with the classification of cells based on their best stimulus. For example, all but 1 of the 21 cells in the N groups were NaCl best; all of the sucrose best cells were linked as the S group and two cells that were quinine best formed the quinine group. Among cells in the more broadly tuned groups, NH1-2 and H, 19 of 29 (66%) were mixture best.

**Table 2A.** Number of NTS cells that show increases or decreases in their responses to mixtures with respect to the MEC

<table>
<thead>
<tr>
<th>Best Stimulus</th>
<th>NH</th>
<th>NS</th>
<th>NQ</th>
<th>HS</th>
<th>HQ</th>
<th>SQ</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>&gt;MEC</td>
<td>&lt;MEC</td>
<td>&gt;MEC</td>
<td>&lt;MEC</td>
<td>&gt;MEC</td>
<td>&lt;MEC</td>
</tr>
<tr>
<td>N (28)</td>
<td>0.68 ± 0.03</td>
<td>0</td>
<td>12</td>
<td>0</td>
<td>9</td>
<td>1</td>
</tr>
<tr>
<td>H (3)</td>
<td>0.76 ± 0.03</td>
<td>0</td>
<td>41</td>
<td>0</td>
<td>31</td>
<td>3</td>
</tr>
<tr>
<td>NH (11)</td>
<td>0.79 ± 0.02</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>33</td>
<td>0</td>
</tr>
<tr>
<td>HS (5)</td>
<td>0.85 ± 0.05</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>9</td>
<td>18</td>
</tr>
<tr>
<td>S (4)</td>
<td>0.63 ± 0.06</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>25</td>
<td>25</td>
</tr>
<tr>
<td>NQ (3)</td>
<td>0.60 ± 0.16</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>20</td>
<td>20</td>
</tr>
<tr>
<td>NS (2)</td>
<td>0.86</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

Cells are grouped according to their best stimulus. Bottom set of numbers are percentages. Number of cells is in parentheses. NTS, nucleus of the solitary tract; MEC, more effective component.

**Table 2B.** Number of NTS cells that show increases or decreases in their responses to mixtures with respect to the sum of the responses to the components

<table>
<thead>
<tr>
<th>Best Stimulus</th>
<th>NH</th>
<th>NS</th>
<th>NQ</th>
<th>HS</th>
<th>HQ</th>
<th>SQ</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>&gt;SUM</td>
<td>&lt;SUM</td>
<td>&gt;SUM</td>
<td>&lt;SUM</td>
<td>&gt;SUM</td>
<td>&lt;SUM</td>
</tr>
<tr>
<td>N (28)</td>
<td>0</td>
<td>24</td>
<td>0</td>
<td>13</td>
<td>0</td>
<td>20</td>
</tr>
<tr>
<td>H (3)</td>
<td>0</td>
<td>86</td>
<td>0</td>
<td>46</td>
<td>0</td>
<td>71</td>
</tr>
<tr>
<td>NH (11)</td>
<td>0</td>
<td>3</td>
<td>0</td>
<td>1</td>
<td>0</td>
<td>2</td>
</tr>
<tr>
<td>HS (5)</td>
<td>0</td>
<td>100</td>
<td>0</td>
<td>33</td>
<td>0</td>
<td>67</td>
</tr>
<tr>
<td>S (4)</td>
<td>1</td>
<td>5</td>
<td>0</td>
<td>4</td>
<td>0</td>
<td>5</td>
</tr>
<tr>
<td>NQ (3)</td>
<td>9</td>
<td>45</td>
<td>0</td>
<td>38</td>
<td>0</td>
<td>45</td>
</tr>
<tr>
<td>NS (2)</td>
<td>0</td>
<td>60</td>
<td>0</td>
<td>60</td>
<td>0</td>
<td>80</td>
</tr>
</tbody>
</table>

Cells are grouped according to their best stimulus.
relationships contained in Table 3A. MDS analyses then positioned each taste stimulus in a three-dimensional “taste space” where similar across neuron patterns were close together and dissimilar patterns were far apart. Results, presented in Fig. 8, showed that across neuron responses to S, N, H, and Q were widely separated from each other while across neuron responses to HS, SQ, and HQ were placed roughly between the across neuron responses evoked by their component tastants. Distances from each of the taste mixtures to all single-component tastants were calculated using the coordinates of the MDS taste space. Table 3B shows the results of this analysis. It can be seen that with the exception of HQ, all taste mixtures were placed closest to their MECs. For mixtures of H and Q, the suppression of the response to HCl by the addition of quinine resulted in the placement of HQ nearer to quinine than to HCl.

Two further analyses showed that with repetition of stimulus presentation, some cells shift their relative sensitivity across tastants, but the across-neuron patterns remain similar. First, responses to all tastants from the first, third, and fifth blocks of trials across cells were treated as though each response profile were derived from a different cell. There were 36 cells for which five replications of all 10 tastants were available. A hierarchical cluster analysis was then used to determine whether replicate response profiles from the same cell would be placed in clusters with similar sensitivity across taste stim-
The dendrograms that resulted from these analyses are shown in Fig. 9 for trials 1, 3, and 5. It is apparent that cluster analyses from each set of trials suggested groups of responses profiles that were similar in each set of trials, i.e., N, H, NH, and S groups. This result is similar to that shown in Fig. 7 using mean response rates from all 56 cells. Average response profiles from cells in the N, NH, and H groups, shown in Fig. 10, are also similar to those in Fig. 7. Among the 36 cells included in these analyses, 11 (31%) were members of different cell groups depending on the particular block of trials. This included 7 of the 13 mixture-best cells in the group (54%) and 3 of the 26 single-component-best cells in the group (12%). With one exception, all “shifts” were among the N, H, and NH groups. A second cluster analysis was applied to the ANPs of response for each stimulus recorded in trials 1, 3, and 5; results are shown in Fig. 11. Note that the cluster analysis in Fig. 11 grouped taste stimuli rather than individual cells, as in Fig. 9. It can be seen that in every case, the ANPs for all three trials were always identified as more similar to each other than to any trial of any other stimulus.
TABLE 3B. Distances in arbitrary units of each taste mixture from single-component stimuli in the MDS space

<table>
<thead>
<tr>
<th></th>
<th>NH</th>
<th>NS</th>
<th>NQ</th>
<th>HS</th>
<th>HQ</th>
<th>SQ</th>
</tr>
</thead>
<tbody>
<tr>
<td>S</td>
<td>2.44</td>
<td>2.21</td>
<td>2.33</td>
<td>1.83</td>
<td>2.17</td>
<td>0.99</td>
</tr>
<tr>
<td>N</td>
<td>0.58</td>
<td>0.29</td>
<td>0.27</td>
<td>1.56</td>
<td>1.07</td>
<td>1.50</td>
</tr>
<tr>
<td>H</td>
<td>0.93</td>
<td>1.61</td>
<td>1.48</td>
<td>0.81</td>
<td>1.03</td>
<td>1.78</td>
</tr>
<tr>
<td>Q</td>
<td>1.34</td>
<td>1.61</td>
<td>1.36</td>
<td>1.49</td>
<td>0.67</td>
<td>1.93</td>
</tr>
</tbody>
</table>

**Discussion**

Electrophysiological responses to NaCl, HCl, quinine, and sucrose and to their undiluted binary mixtures were recorded in 56 cells in the NTS of anesthetized rats. In most cells, responses to taste mixtures were highly correlated and close in magnitude to the responses to the MEC of the mixture. However, mixture suppression and enhancement for some subset of mixtures was also observed in most every cell. The best stimulus of a cell was a good predictor of its pattern of mixture suppression and/or enhancement across tastants in many ways mirroring similar effects described in peripheral afferents (Frank 1989). In addition to cell types that responded best to one of the four single-component tastants, groups of cells that consistently responded best to particular taste mixtures were described. These mixture-best cells were more broadly tuned than were cells that responded best to single-component stimuli and showed evidence of convergence of sensitivity from more than one taste quality. Functionally, these cells may amplify the neural signal produced by unique configurations of basic taste qualities.

In 36 cells all 10 tastants were presented between 5 and 23 times. Consistent with previous reports (Di Lorenzo and Victor 2003, 2007), response magnitude varied considerably across trials; when responses to the four single-component taste stimuli were considered, only 61% of these cells showed a consistent best single-component stimulus. If it is assumed that the best stimulus identifies the message that is conveyed by a given cell, then these results imply that on a trial-by-trial basis response magnitude alone produces ambiguous signals. Moreover, for most cells, when responses to mixtures were considered along with responses to single-component tastants, the variability of response magnitudes across blocks of trials underscored the difficulty in discriminating any stimulus from the array. This was evidenced by similar responses magnitudes to multiple stimuli and by changes in cluster membership when responses from different blocks of trials were considered. On the other hand, the ANPs of responses to the same stimulus across different blocks of trials were more similar to each other than to those of other stimuli (Fig. 11). These results suggest that ANPs convey a more consistent and reliable message about interstimulus differences that do the responses from individual cells.

Trial-to-trial variability of NTS taste responses has been described in previous studies (Di Lorenzo and Victor 2003, 2007; Roussin et al. 2008). While the source of this variability may represent noise (random variability) in the system, it is also possible that these cells are tracking some endogenous variable such as attentional state, blood-glucose level, etc. We did not address this issue in the present study, but we are aware that some underlying factor may account for response variability.

Peripheral versus central nervous system (CNS) processing of taste mixtures

Reports of responses to taste mixtures in the CT nerve showed that the effects of taste mixtures varied according to the best stimulus classification (Frank 1989; Hyman and Frank 1980b); present results suggest that this was also true to a large extent in the NTS. For example, Frank (1989) reported that responses to a CT fiber’s best stimulus were suppressed when that stimulus was mixed with a nonbest stimulus in “specialist” fibers, i.e., narrowly tuned, that were N best or S best; this was also true of N- or S-best cells in the NTS of the rat. As is evident in Table 2, the great majority of N- and S-best cells showed responses to N-containing mixtures that were either unaffected or significantly less than the response to their best stimulus presented alone. (Parenthetically, these effects are not seen if one reclassifies mixture-best cells by their best single-component stimulus, e.g., an NH-best cell might become an N-best cell.) Frank (1989) also stated that “generalist” fibers, i.e., broadly tuned, usually H best, were always enhanced when H was mixed with another stimulus. In general agreement with this observation, in rat NTS, if one counts all three H-best cells along with all 21 mixture-best cells as analogous to generalist CT fibers, then mixture enhancement was seen in responses to NH in 15 of 24 cells (61%) and to HS in 10 cells (41%), but mixture suppression was seen for responses to HQ in 14 cells (58%). Similar findings have been reported in the parabrachial nucleus of the pons (PbN, the main target of taste-related NTS projections) in the hamster (Travers and Smith 1984; Vogt and Smith 1993a,b).

Studies of mixture effects in the hamster CT (Frank et al. 2005; Hyman and Frank 1980b) and PbN (Vogt and Smith 1993a) have consistently noted the suppression of sucrose by quinine. This was also seen in the rat NTS, though mixture...
enhancement was also observed. Vogt and Smith (1993a) reported that the degree of suppression was correlated with sensitivity to sucrose, but this was not true in the present study. Because these and other mixture effects are known to be dependent on stimulus intensity (Hyman and Frank 1980a,b; Vogt and Smith 1993a,b), discrepancies may reflect the fact that only one concentration of S and Q was used in the present study.

Discrepancies between the reported effects of taste mixtures in the CT and the NTS may reflect species differences (most CT studies were done in hamster) but may also be the result of the convergence of multiple inputs to NTS cells. For example, in addition to the CT nerve, taste-related input to NTS cells also originates in the glossopharyngeal (Frank 1991) and greater superficial petrosal (Travers et al. 1986) nerves. Centrifugal input also modulates taste responses in this area (e.g., Di Lorenzo and Monroe 1995; Smith and Li 2000), and there is a rich network of local interneurons (Beckman and Whitehead 1991). Finally, many taste-responsive NTS cells also respond to thermal (Ogawa et al. 1988) and tactile (Ogawa et al. 1984) input, and these modalities may also modulate NTS responses.

Commonalities among the present results and those of previous studies of the hamster PbN (Travers and Smith 1984; Vogt and Smith 1993a,b, 1994) suggest that taste mixtures are processed similarly in brain stem nuclei across species. For example, in both rat NTS and the hamster PbN, most responses to taste mixtures were not significantly different from the responses to the MEC of the mixture. Additionally, mixture responses in both rat NTS and hamster PbN showed mixture enhancement (~15%) or suppression (~15–25%) in about the same proportion of responses (Travers and Smith 1984). The further observation that responses to mixtures were most strongly correlated with the responses to the components of the mixture suggests that taste qualities contained in a mixture are identifiable within that mixture and, conversely, that taste mixtures only evoke the taste qualities of the components (Travers and Smith 1984). Behavioral studies using conditioned taste aversion support this contention (Formaker and Frank 1996; Nowlis and Frank 1981; Smith and Theodore 1984).

In the present study, 36% of NTS cells that did not respond to a given stimulus showed responses that were affected by that

![FIG. 8. Results of MDS analyses in 3 dimensions for all taste stimuli based on Pearson product-moment correlations. Responses from all 56 cells were used in this analysis. For cells where a given stimulus was repeated, the average response across trials was used. Stress values were as follows: for 1 dimension, 0.28569, \( R^2 = 0.89132 \); for 2 dimensions, 0.11737, \( R^2 = 0.96995 \); for 3 dimensions, 0.02740, \( R^2 = 0.99842 \); for 4 dimensions, 0.00621, \( R^2 = 0.99992 \); for 5 dimensions, 0.00744, \( R^2 = 0.99995 \).](http://jn.physiology.org/)

![FIG. 9. Results of hierarchical cluster analyses for all cells for trials 1, 3, and 5 of all stimuli; each block consists of a single presentation of all 10 stimuli. Responses from 36 cells were used in this analysis. Response profiles from each block of trials in each cell are grouped according to their similarity as measured by the Pearson product-moment correlation. Clusters are joined at distances that are proportional to their intercluster correlations. Clusters are labeled as in Fig. 7. See text for details.](http://jn.physiology.org/)
stimulus when it was a component of a mixture. This result suggests that these cells receive input associated with that stimulus, even when this input does not by itself produce an increase in firing rate. Such phenomena have also been documented in peripheral nerves (e.g., Sakurai et al. 2000), suggesting that interactions at the level of the taste receptor cell and/or taste bud may be at play. Alternatively, these effects may be the result of the interrelationships of excitatory and inhibitory inputs within the NTS. A better understanding of NTS circuitry will be necessary for a more complete interpretation of these effects.

Role for mixture-best cells in neural coding of complex taste stimuli

Present results provided strong evidence that the rat NTS contains cells that respond more vigorously to a taste mixture than to any single-component tastant, i.e., mixture-best cells. Travers and Smith (1984) also noted the presence of mixture-best cells in the PbN of the hamster. In both the NTS and PbN, 30–40% of the taste responsive cells were mixture best and showed responses that were 1.1–2.1 times the response to the best single-component tastant. Travers and Smith (1984) concluded that these cells did not constitute a unique cell type due in large part to the close association of the responses to the best mixture and the responses to the best single-component stimulus. That is, they emphasized the fact that the great majority of mixture-best PbN cells responded best to mixtures containing their best single-component stimulus and therefore did not signal any unique stimulus. This relationship between best single-component and best mixture was also seen in mixture-best NTS cells; however, in the present study, data from many stimulus replications showed that the best single-component stimulus in mixture-best cells often varied across blocks of stimulus presentations, but the best mixture did not. In addition, present data showed that mixture-best cells were significantly more broadly tuned than single-component best cells and showed evidence of sensitivity to more than one taste quality. For example, NH-best cells, identified by cluster analyses, responded well to both N and H as well as mixtures containing N or H, whereas the N group only responded well to N and mixtures containing N, and the H group only responded well to N, H, and mixtures containing H. The functional characteristics of mixture-best cells bolster the argument that they form a separate class of cell and further suggest that they may be specialized to signal unique configurations of tastants.

The issue of cell types in the gustatory system has been a thorny one for several decades. Although people have tried to apply the best stimulus classification scheme to cells in the CNS, results are not as clear as they have been in the peripheral nervous system (see Lemon and Smith 2006). Taste-responsive cells in the CNS are more broadly tuned than are peripheral nerve fibers and response variability across trials makes classification based on a best stimulus...
problematical (Di Lorenzo and Victor 2003; 2007). Even so, if one argues that each class of incoming fibers, grouped according to their best stimulus, represents an information “channel” for a single taste quality (Chandrashekar et al. 2006), then the presentation of a mixture of tastants of two different qualities should logically excite two (independent) channels. Extending this view to the more broadly tuned cells in the CNS, one may conclude that either the sideband responses (defined as responses to taste qualities other than that of the best stimulus for a cell) are noise (in which case the CNS is very noisy) or that CNS cells generally contribute information about more than one taste quality category. This conundrum is amplified when the system is presented with complex tastants such as mixtures. Viewed with the argument that sideband responses are noise, mixtures would excite more cells than would any single component stimulus (by definition) but would also generate the most noise in the system because more cells with relevant sideband sensitivity would also respond. Thus mixtures would be less identifiable than single component tastants. However, behavioral data show that conditioned aversions to taste mixtures are stronger than the generalization of those aversions to the component tastants (Frank 1989; Nowlis and Frank 1981), suggesting that the mixture has an identifiable property that is separable from its components. In addition, Frank (1989) showed that the components of mixtures containing NaCl and sucrose were more difficult for hamsters to identify than the mixture themselves following conditioned aversion training.

From the alternative view that CNS cells can convey information about more than one taste quality, it is difficult to see how the firing rate in response to a tantant by itself can convey enough information to identify a taste stimulus, especially if that stimulus is a taste mixture (see Lemon and Smith 2006). While present data show that there are groups of cells that respond best to some taste mixtures and that show evidence of convergence of sensitivities to the basic taste qualities (see Fig. 7), it seems unlikely that these cells play the same role in encoding mixtures as their best stimulus counterparts in the peripheral nervous system. Instead other coding mechanisms such as across neuron patterns or the temporal characteristics of their responses may add crucial information (see Di Lorenzo and Victor 2003, 2007; Katz et al. 2001, 2002; Roussin et al. 2008). In the present study, we demonstrated that across neuron patterns of responses produced a logical map of the taste space (Fig. 8) and that these patterns were tightly associated despite trial-to trial variability (Fig. 11). Further, we showed that mixture-best cells enhanced the contrast among taste mixtures within the context of an across neuron pattern. These data are in agreement with previous analyses of hamster NTS responses showing that the cells that are most responsive to a given tantant exert the greatest influence on the ANP (Smith et al. 1983). The contribution of temporal coding to the identification of taste mixtures was not addressed in the present study. However, previous reports suggest that this mechanism may make an important contribution to the discrimination of taste mixtures (Di Lorenzo and Victor 2003, 2007; Roussin et al. 2008).

**Analytical nature of taste**

Data from behavioral experiments have shown that both rats and hamsters are capable of identifying the components of a taste mixture but are also consistent with the idea that mixtures may represent unique and identifiable perceptual entities. For example, conditioned taste aversions to single-component taste stimuli have been shown to generalize to two-component mixtures in proportion to the concentration of each component within that mixture (Smith and Theodore 1984). Conversely, conditioned aversions to taste mixtures generalize specifically to the components of the mixture (Formaker and Frank 1996; Nowlis and Frank 1981). Taken together these data imply that the taste system is analytic and that taste mixtures do not evoke any taste qualities other than those of the components of the mixture. Present data on taste responses in the NTS are compatible with this notion. However, there are also data suggesting that complex tastes may have characteristics that are separate from their components. That is, complex tastants, such as mixtures, may be conceptualized as evoking “configural” taste percepts that may be distinctive enough to have their own identity (see Erickson 2000; Erickson et al. 1990). A good example of such a taste stimulus is ethanol. Conditioned aversions to ethanol have been shown to generalize to a mixture of sucrose and quinine but not to either component alone (Di Lorenzo et al. 1986; Kiefer and Mahadevan 1993).

From an evolutionary standpoint, it makes sense that the taste system would be able to identify unique configurations of taste qualities, especially given the evidence that there are very few taste qualities. Consider for example, the case where an animal becomes ill after eating a food that tastes sweet and sour. Assuming that a conditioned taste aversion is formed to that food, the animal will avoid eating sweet-sour foods the next time they are presented. However, if the association is only to the components of the taste mixture, and not to the combination (configuration) of tastes, then all sweet and all sour foods will be avoided. It is not difficult to imagine that with further experiential learning, all foods will be avoided because all foods would contain at least one component of the conditioned stimuli. In contrast, if an animal could learn to avoid only the sweet-sour configuration, then sweet-only and sour-only foods would remain acceptable to some degree. It is not surprising then that conditioned aversions to taste mixtures are always stronger to the mixture than to the components (Frank 1989; Nowlis and Frank 1981). The observation that there are groups of cells in the NTS that respond preferentially to certain mixtures may provide the neurophysiological basis for configural perception of taste and, further, suggests that this type of perception occurs at the earliest level of central processing of taste information.

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**References**
