late response component. Removing the front part of the call caused the early response component to drop out, without further affecting the late part of the response (Fig. 4D). When an additional small piece of the call (35 msec in duration) was deleted, the late response component also disappeared (Fig. 4E). Next, a 135-msec piece containing this 35-msec portion was replaced (Fig. 4-F). However, the late response component was only partially restored (compare Fig. 4, D–F). Thus, while a limited portion of this call was necessary for the production of the later response components, it was not sufficient to produce a full-strength response. It was only through interactions with other parts of the call, which did not produce separate response components in this cell, that a strong late response occurred.

The responses of cell I to vocalizations do not seem to be based on the analysis of simple acoustic cues but on analysis of the temporal structure of the calls. Neurons of this type permit the auditory cortex to code many different temporal patterns and are likely to be the elements underlying the cortex's demonstrated role in temporal pattern analysis (*1*). Conceivably, interpretation of specific vocalizations requires neurons with low noise and high response reliability to the total acoustic structure of a single call. Cell II comes close to meeting these criteria. It responded with high probability only to the Isolation Peep and had slow spontaneous activity, and several parts of the call were coded in the relatively simple response pattern. However, two factors imply that this cell is not at the highest level of specificity. First, it continued to respond to incomplete versions of the Isolation Peep, and second, it responded, although with much lower probability, to other vocalizations.

Our results support the suggestion that the auditory cortex is concerned with the analysis of complex auditory signals. They also suggest that, at least for the squirrel monkey, this structure plays an important role in the coding and interpretation of sounds used in species-specific communication.

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5. A magnetic tape was made with each vocalization represented 25 times, 4.5 sec intervening between successive presentations. A pulse on the second channel of the tape preceded the onset of each vocalization; it triggered the instrumentation for data display and data analysis. The other stimuli were repeated at 2-sec intervals. The duration of tone and noise bursts was 300 msec. Clicks were produced by 0.3 msec rectangular pulses. Peak intensity for the vocalizations was 60 to 80 db relative to 0.0002 dyne/cm^2. Intensities of the tone and noise bursts were also within this range.
6. We define "response complexity" by subjective appraisal of the dot displays: a response is *simple* if it consists of up to three visibly separable columns of dots; *complex* when it has more than that.
7. J. D. Newman and Z. Wollberg, in preparation.
8. Our code numbers for cells I and II are, respectively: "CLG II-0.5,4" and "CRH I-1.5,1."
9. The Isolation Peep, although it is the longest call, has one of the simplest acoustic structures among calls we used (*3*).
10. We thank P. G. Nelson, D. Symmes, G. Fischbach, and H. Levitan for their comments.

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Human Taste Papilla Stimulation: Stability of Quality Judgments over Time

Abstract. *Human taste papillae are sensitive to two or more chemical stimuli. Individual papillae produce stable response patterns (quality and intensity) over a month's time. Hence, the response pattern does not appear to be affected by turnover of receptor cells.*

Studies of taste experience after chemical stimulation of human taste papillae have left a confusing picture of papillae response properties. Von Bekesy reported that each single taste papilla mediates only one taste quality (*1*). But, Harper *et al.* found that a taste papilla generally mediates more than one quality (*2*); they criticized the von Bekesy experiment on the grounds that there was no control for spread of stimulus to neighboring papillae and that there was no estimate of the reliability of data. To handle these problems they used a tiny suction ring which confined the stimuli to a given papilla, and they employed a double-blind design with statistical treatment of the data. However, their procedure appears to have produced a great loss of sensitivity because most of the taste responses were faint and ambiguous, even with very strong stimuli (*3*). Their suction ring may have produced enough tactile interference to diminish the subjects' sensitivity. We decided to reexamine the response properties of taste papillae by using less disturbing stimulation procedures. We employed essentially von Bekesy's technique of stimulation but included a control for stimulus spread, a double-blind design, and statistical estimates of data reliability.

In addition, we wanted to know if the response properties of a single papilla were stable over time. Beidler and Smallman (*4*) found that rat taste receptors have a life of about 250 hours; new receptors appear to be differentiated from surrounding cells. If human taste cells turn over at the same rate, then a taste papilla would have several new sets of receptors over a month's period (*5*). It is possible that a new set of receptors would mediate a different pattern of taste activity in the papilla.

Two subjects (1 male, 30 years; 1 female, 21 years) were given a preliminary test in making whole-mouth taste discriminations. This test consisted of recognition of sweet (0.04*M* sucrose), sour (0.001*M* citric acid), salty (0.04*M* sodium chloride), bitter ($3 \times 10^{-4}M$ quinine sulfate), and tap water. Each stimulus appeared five times in a randomly mixed order. After sampling 20 cm^3 of solution, the subjects reported the quality, rinsed their mouths with 20 cm^3 of water, and rested 1 minute until the next trial. As was expected with these concentrations, each taste report was the characteristic quality for that solution. Water yielded various weak taste sensations or none at all. All solutions were made with distilled water and were used at room temperature. Citric acid, sodium chloride, and quinine sulfate were reagent grade, and sucrose

was commercial grade. Quinine sulfate was prepared as a stock solution because of solubility difficulty. All other solutions were prepared fresh each test day.

The subjects were then seated in front of a special microscope stand which was equipped with a chin rest for the subject and an arm rest for the experimenter. Taste papillae were located and magnified 15 times with a binocular dissecting microscope. A map was made of the fungiform papillae located in approximately a 1 cm^2 region on the dorsal surface of the anterior 3 cm of tongue. Solutions were delivered by a variable-speed infusion pump, connected by PE 10 tubing to 30-gage stainless-steel tubing; the steel tubes were mounted in small acrylic plastic holders for ease of manipulation.

With a little practice, it was easy to position the tip of the delivery tube over a selected papilla. Drop size was controlled by pumping speed and duration (automatically controlled by an electronic interval timer in series with the pump motor), and size was selected to just cover the top surface of the papilla (about 0.1 μl) (*6*). No attempt was made to disturb the position of the papilla. Just before stimulation, the subject extended his tongue to expose about 3 cm of the front part. Within 5 seconds after stimulation, the subject was required to choose one or more of five possible responses ("sweet", "sour", "bitter", "salty", and "no taste"), before he retracted his tongue. The subject responded by pointing to the appropriate label on the arm of the scope stand. After responding, the subject rinsed with approximately 20 cm^3 of water and then expectorated. Then the subject rated the intensity of the taste on a 5-point scale (1, weak; 5, strong) and rested for 1 minute between stimulations. No discomfort was reported during the testing.

Single papilla training in one subject (J.F.) was begun by stimulating fungiform papillae on the front of the tongue with 0.4*M* sucrose until two papillae were found which yielded a sweet sensation. For each papilla, water and 0.4*M* sucrose were each presented five times in a mixed order. The experimenter informed the subject of the stimulus after each response. This subject made only one "mistake" over these trials. The other subject (C.L.) did not receive this part of the training. Both subjects received single papilla training

Table 1. Taste quality for papillae from each of two subjects: ratio of the number of test days on which criterion was reached to the total number of test days. Criterion was defined as the same quality response to two of three presentations of the same chemical stimulus.

Taste quality	Response ratio					
	Sucrose	NaCl	Citric acid	Quinine sulfate	Water	Control
			Subject J.F., papilla 1			
Sweet	6/6*†					
Salt						
Sour		5/6†	6/6*†		2/6	
Bitter	2/6			2/6	2/6	
None		1/6		3/6†	1/6	6/6*†
			Subject C.L., papilla 1			
Sweet	5/6*†			1/6		1/6
Salt						
Sour	1/6	6/6*	5/6*†	3/6†	2/6	
Bitter						
None				1/6	4/6†	4/6*†
			Subject J.F., papilla 2			
Sweet	5/6*†					
Salt		2/6	3/6			
Sour		4/6	4/6*			
Bitter				1/6	2/6	
None	1/6			5/6†	4/6	5/6*†
			Subject C.L., papilla 2‡			
Sweet	3/4*†					
Salt		1/4	2/4			1/4
Sour	1/4	3/4	2/4	1/4		
Bitter						
None				3/4†	3/4	

* Chi-square test significant for response appropriate to stimulus; $P < .01$. † Criterion stability: criterion response over 2-week test period (three successive tests). Note that some stimuli are given double responses. For example, J.F. responded to sucrose applied to papilla 1 with sweet-bitter on several trials. ‡ Subject ill on two test days.

stimulation for 15 trials per day for 2 days with a randomly mixed order of citric acid (0.1*M*), sodium chloride (0.4*M*), sucrose (0.4*M*), quinine sulfate ($3 \times 10^{-4}M$), and water. After each response, the experimenter informed the subject about the stimulus chemical but did not give a "correct" category label. The subjects were told that their response was accepted as correct and that they were not to be concerned about giving a "typical" response. Thus we trained the subjects to detect differences in tastes produced by the different stimuli and to report these differences consistently.

Each test consisted of three presentations for each of the five stimuli. Five control trials utilizing an effective stimulus were delivered within 1 mm next

Table 2. Taste intensity: ratio of the number of test days on which criterion was reached to the total number of test days. Same subjects, same papillae, same definition of criterion as in Table 1.

Taste Intensity	Repsonse ratio				
	Sucrose	NaCl	Citric acid	Quinine sulfate	Water
			Subject J.F., papilla 1		
Weak	4/6*	1/6	1/6	2/6	2/6
Medium	2/6	3/6	3/6	1/6	
Strong		1/6	2/6		2/6
			Subject C.L., papilla 1		
Weak		1/6		2/6	
Medium	6/6*	3/6	3/6	2/6	1/6
Strong			3/6		
			Subect J.F., papilla 2		
Weak	4/6*	2/6	4/6*	1/6	2/6
Medium	1/6	2/6	2/6		
Strong		2/6			
			Subject C.L., papilla 2†		
Weak	1/4				
Medium		4/4*	3/4*	1/4	
Strong					

* Criterion stability: criterion response for at least two-thirds of total test days. † Subject ill on two test days.

to the papilla. Each control stimulation was delivered to a different location systematically varied in a clock-wise fashion around the tested papilla. In total, 20 stimulus trials were presented in a randomly mixed order. In order to insure against experimenter bias, stimuli were coded by someone else so that on any trial the experimenter did not know what solution was being presented. Stability of response patterns of papillae over time was examined by testing both papillae once a week for 3 weeks or more. Each papilla had its own random presentation order which was fixed over the test period. Testing was always done at the same time over the test period.

Criterion for stability of response quality was defined as the choice of the same response quality (with an intensity rating of 1 or better) for two out of three presentations of the same chemical for three consecutive tests (2 weeks). Criterion stability of response intensity was defined as the choice of the same range of response intensity (1 to 2, weak; 2 to 4, medium; 4 to 5, strong for two out of three presentations of the same chemical for at least two-thirds (2 to 3 weeks) of the total number of tests. Overall reliability of relating a specific quality to a particular chemical was determined by a chi-square test (*7*).

Sucrose gave a stable "sweet" response in both papillae for J.F. and one papilla for C.L. (Table 1). In addition, sodium chloride gave stable "sour" responses in one papilla for both subjects. Although the two subjects occasionally gave "salty" responses to sodium chloride stimulation, the predominant response was "sour". In the other two papillae, citric acid yielded primarily "salty" or "sour" responses on different test days. Citric acid gave stable "sour" responses in one papilla for both subjects. Water and quinine gave predominantly "no taste," although they yielded a few weak "sour" or "bitter" responses.

Sucrose produced weak to medium sensations (Table 2). Stability of sucrose intensity was seen for two papillae in J.F. and one papilla in C.L. Citric acid gave stable intensity responses for one papilla per subject. These response intensities ranged from weak to strong in both subjects. Sodium chloride response range was from weak to strong in one subject and from weak to medium in the other subject. Criterion intensity stability for sodium chloride was reached in one papilla. Results for quinine sulfate were generally inconsistent. On many trials stimulation produced "no taste." On some it produced a "bitter" or "sour" response. In general it was about as effective a stimulus as water.

Control stimulation reliably produced a "no taste" response for three out of four papillae. Sometimes there was a taste response which was characteristic for the stimulus used. This was probably due to the proximity of the test papilla to a neighboring active papilla. For example, papilla 2 of J.F. lay about 1 mm from the neighboring active papilla at one control site. Hence, it was virtually impossible to avoid stimulating the adjacent papilla in that off-papilla control position. Control stimulation delivered to the other sites around the test papilla did not produce taste sensations, even though the stimulus drop was delivered equidistant from the test papilla in all positions. It is probable, therefore, that spread of stimulation from the test papilla was of no consequence in this study.

These results show that the single fungiform taste papilla is capable of responding to both sucrose and citric acid with the typical distinctive tastes and that the response pattern is stable over a month's test. As indicated in previous work (*8*) the single papilla can respond to more than two taste substances with the appropriate distinctive tastes. This finding is in apparent contradiction to the results of von Bekesy. It must be remembered, however, that von Bekesy used threshold concentrations of taste substances, whereas the contradictory experiments used suprathreshold strengths. Our failure to obtain the reliable "salty" responses to a strong concentration of NaCl is puzzling. It is possible that simultaneous stimulation of several papillae will be necessary to obtain a clear "salty" response.

Neither of the current theories of taste coding (specific taste fibers versus specific across-fiber pattern) are supported by our finding of stable response patterns over a month-long test. It is known that most, if not all, individual rat taste bud cells have a short life and are continuously replaced by differentiation of nearby cells. Further, it has been proposed that the innervation of any particular type of taste receptor in the rat is a random event (*9*). At first glance, the problem of constructing a stable analyzer out of such an unstable system of transducers seems formidable (*10*). However, since many receptors mediate a single papilla taste experience, it is possible that there are a sufficient number of sugar or acid receptors present at any given time to yield the typical response. Another answer may be that the human papilla is not as labile a system as the rat papilla (*11*). We obviously need to learn more about the physiology of the human taste system before we can explain the taste experience mediated by that system.

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5. C. Pfaffmann, in *Handbook of Physiology*, H. W. Magoun, Ed. (American Physiological Society, Washington, D.C., 1959), vol. 1, section 1, p. 507; L. M. Beidler, in *Taste and Smell in Vertebrates*, G. E. W. Wolstenholme and J. Knight, Eds. (Churchill, London, 1970), p. 51. The human fungiform papilla contains three to four taste buds; each bud contains about 50 cells. Not all of these cells within the bud are considered to be active taste receptors.
6. After completion of the experiment, we determined the size of the stimulus drop by comparison with a drop produced by a precision, repeating 10-μl syringe. There was very little variation in drop size from stimualtion to stimulation. Occasionally the stimulus drop would be too small or too large relative to the diameter of the papilla. We made the necessary adjustment by changing pumping time.
7. Chi-square analysis was taken from Harper *et al.* (*2*). Percentage responses expected by chance were calculated from the ratio of the number of times a given quality category was used (for example, "sour") to the total number of presentations of all chemicals. This value was compared with the percentage of obained responses (for example, "sour" to citric acid stimulation).
8. In the Harper *et al.* experiment (*2*) only one subject reliably recognized four different tastes for each of three papillae. The other subject with multiple taste discriminations reliably reported "sweet" to sucrose stimulation and "sour" to citric acid stimulation for each of two papillae.
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