in concentration of serotonin in the brain, presumably as a result of section and degeneration of fibers which normally produce serotonin, also reduces the amount of enzyme available for the synthesis of the amine. Whether the decrease in amine concentration is a result of a reduced synthesis secondary to the loss of 5-HTP decarboxylase activity cannot be stated with certainty, since it is not possible to correlate decreases in enzyme activity as measured in vitro with synthesis of serotonin in vivo. For example, Brodie *et al.* (*9*) have shown that doses of N-(3-hydroxybenzyl)-N-methyl hydrazine, which produce an apparent 75-percent inhibition of 5-HTP decarboxylase as measured in vitro, do not affect the biosynthesis of brain serotonin in vivo. The demonstration by Grahame-Smith (*10*) of the enzymatic hydroxylation of tryptophan in brain homogenates must also be considered. A determination of the effect of lesions of the medial forebrain bundle on this first step in the biosynthesis of serotonin would be helpful in deciding whether such lesions lower the concentrations of serotonin by reducing one or both of the enzymatic activities necessary for the synthesis of the amine. Despite these considerations, it is interesting that the percentage decrease in 5-HTP decarboxylase activity is only about half the percentage reduction in amine concentration. This quantitative difference in percentage change of amine and enzyme suggests that part of the decrease in amine concentration may be secondary to some non-enzymatic mechanism such as loss of binding sites due to degeneration of the fiber tract. The failure of either lesion to alter malic dehydrogenase activity indicates that the change in 5-HTP decarboxylase is not a reflection of some non-specific effect of the lesion on cellular enzymatic activity.

The similarity of the effect of sectioning the medial forebrain bundle on the concentration of serotonin in the brain to the effect of sectioning autonomic nerves on tissue catecholamines and ganglionic acetylcholine has been discussed (*1*, *2*). In addition, Hebb and Waites (*11*) demonstrated that the loss of capacity of the superior cervical ganglion to synthesize acetylcholine after section of the cervical sympathetic trunk (*12*) was due to a decrease in choline acetylase activity in the ganglion. Similar effects on dihydroxyphenylalanine decarboxylase in spinal cord and sympathetic nerve have been presented by Anden *et al.* (*13*). Thus, our findings demonstrating a decrease in both serotonin and 5-HTP decarboxylase after section of the medial forebrain bundle are consistent with the suggestion that the bundle contains serotonin-producing fibers. We have recently shown, however, that the decrease in amine occurs both in areas directly innervated by the medial forebrain bundle and in areas related to it only through polysynaptic connections (*8*). The reduction in amine content of these latter areas indicates that the lesion can produce transynaptic neurochemical effects in neurons outside the medial forebrain bundle. The results, therefore, not only demonstrate that the integrity of the medial forebrain bundle is necessary for the maintenance of normal serotonin content and 5-HTP decarboxylase activity in the rat telencephalon, but, in addition, support the view that the brain contains serotonin-producing fibers either within the medial forebrain bundle or under its influence.

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Duplication of Evoked Potential Waveform by Curve of Probability of Firing of a Single Cell

Abstract. *Computer compilation of the probability of firing of a single cell in cat cortex following a physiological sensory stimulus (somatic or light flash) indicates that the frequency distribution of the firing of a single cell closely corresponds to the average waveform of the evoked potential recorded from the same microelectrode. This high correlation holds for both positive and negative and early and late components of the evoked response.*

The relation between evoked potentials and the electrical activity of single cells in brain is still not well understood, and sustained interest and a considerable amount of data have resulted in a number of contrasting points of view (*1*). Most interpretations of this relation have been based on single and multiple superimposed oscilloscope traces, but such small variable samples can yield only impressions of the pattern of single-cell firing and make agreement regarding the relation to the evoked potential difficult to reach. Although the use of nonphysiological (electrical) stimulation to produce synchronously evoked potentials provides better control of the stimulus and response and reduces the variability of the relationship, the probabilistic nature of the relation between the evoked potential and the single cell, which is characteristic of physiologically evoked responses, makes interpretation of oscilloscope data difficult.

Despite frequent disagreement as to which wave components of an evoked potential are attributable to which level of the cortex or cellular mechanism, it is generally agreed that different neural elements sequentially activated relate to the sequential appearance of various components of the waveform of the electrically evoked potential. By means of a computer we have attempted to clarify the relation between the probability of firing of single cortical cells and the components of the waveform of the evoked potential recorded from the same microelectrode. In this way, quantitative data from a large number of single oscilloscope sweeps could be stored in the computer and summed while the original time relations were maintained.

If the suggested relation of individ-

ual cell spikes to evoked potentials (concluded from previous studies in which electrical stimulation was used) also applied to asynchronous evoked-potential and single cell responses to physiological stimuli, then individual cells should show high probabilities of firing corresponding uniquely to specific components of the evoked potential complex (unimodal distribution). However, if a high frequency of firing by individual cells does not correspond uniquely to any single component of the evoked potential complex, but corresponds to a number of components (multimodal distribution), then this would suggest that the entire evoked potential complex is related probabilistically to each cell.

Forty cats were operated on under ether anesthesia; they were then curarized and maintained by means of artificial respiration. The exposed tissue margins were treated with procaine and the eyes were treated with atropine regularly. After 3 to 5 hours, spikes from single cells were recorded from a glass microelectrode, filled with KCl (0.5- to 2 μ-tip), placed on visual or somatic sensory cortex. One microelectrode was used to record both the evoked potential and single cell responses, and these two types of response were differentiated by separate amplifiers set at appropriate frequencies.

In addition the spikes from a single cell activated a Schmitt trigger which provided digital input for one channel of a computer (*2*). The single cell spikes recorded were 0.5 mv to 5 mv in amplitude and thus easily isolated from background activity. The output of the Schmitt trigger was constantly monitored on an oscilloscope along with the single cell spikes to ensure that only the single cell spike and no other electrical activity was activating the trigger. The computer was programmed to provide a frequency distribution with respect to time of the total number of single cell responses in each of the 400 consecutive 2.5-msec computer addresses. Stimuli were presented once during each 1-second sweep on the oscilloscope (3 seconds between sweeps) and the responses of single cells in each address were summed over all sweeps to yield the frequency distribution. Since the stimulus occurred after a constant delay from the start of each sweep, and the computer analysis began at the start of each sweep, this procedure provided a frequency distribution composed of the spikes from an individual cell both before (control) and after stimulation.

Evoked potentials recorded from the microelectrode were averaged in the usual analog-to-digital mode of the CAT 400-B, after mechanical destruction (tapping on the microdrive) of the related cell. Observation of the Schmitt trigger monitor while evoked potentials were being averaged indicated that the evoked potential did not contribute to any single cell response count. Both frequency distributions of responses of single cells and the evoked potential averages were photographed from a "slave" oscilloscope and punched out on paper tape for processing by a general purpose computer (*3*). The computer program for the analysis of the data provided both an overall and a dynamic (moving window) cross-correlation for each single cell frequency distribution and evoked potential average recorded from the same microelectrode site.

Data collected from 100 cells which responded to stimulation (light or somatic) and from their related evoked potentials indicate that the probability of firing for any single cell is to a great extent given by the waveform of the evoked potential recorded from the same microelectrode; that is, the function of the best fit to the frequency distribution of response of any cell following a sensory stimulus is given by the evoked potential. Expressed somewhat differently, a single cortical cell, given enough stimulus presentations, will generate a frequency distribution over time which duplicates the entire evoked potential.

The curve in Fig. 1*a* shows the (frequency–time) distribution of the responses of a single visual cortical cell stimulated 4918 times with a 10-μsec light flash. The averaged evoked potential from the same electrode and location, recorded after destruction of the cell which produced the data in Fig. 1*a*, is given in Fig. 1*b* (average of 200 sweeps). The fit of the averaged evoked potential to the distribution of the responses of the single cell is apparent. Most interesting, perhaps, is the consistency with which the single cell frequency distribution duplicates in detail even the very late (500 to 800 msec) components of the evoked potential. Comparison of Figs. 1*c* and 1*d* shows that the probability of firing for another visual cortical cell after 3150

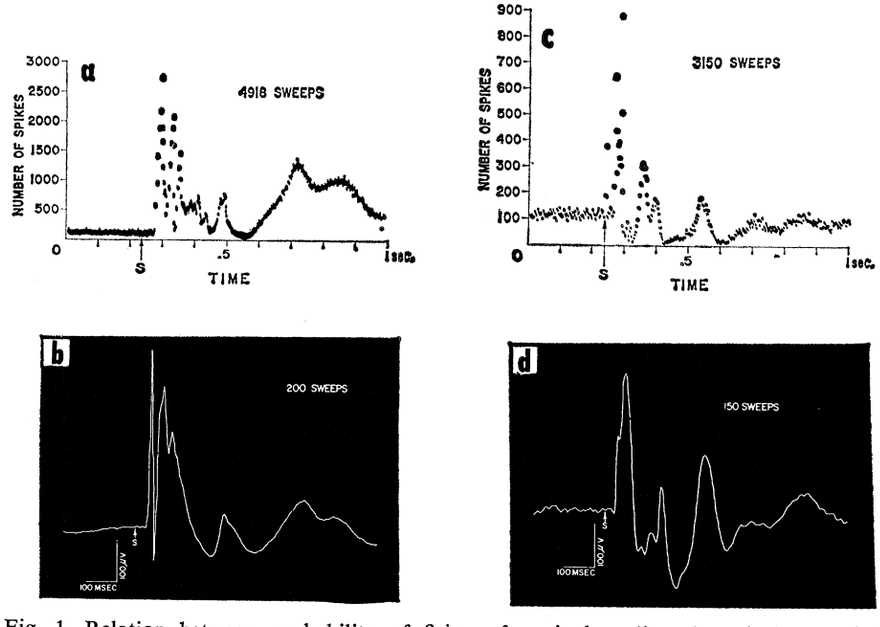

Fig. 1. Relation between probability of firing of a single cell and evoked potential waveform. (*a*) Frequency distribution of spikes from a single cell in the visual cortex of a cat after stimulation with 4918 flashes; (*b*) averaged evoked potential (200 oscilloscope sweeps) recorded from the same microelectrode, after cell death ($r = .60$; $p < .001$). Similarly, spike distribution for a single cell is shown in (*c*) (3150 sweeps) and the corresponding averaged evoked potential in (*d*) (150 sweeps) ($r = .51$; $p < .001$). Ordinate (for unit distributions): number of times the cell fired in response to light flash. Abscissa (for unit distributions): time, in 100-msec divisions.

stimulations with a light flash is accurately reproduced by the shape of the averaged evoked potential (average of 150 sweeps) recorded from the same microelectrode. Since these distributions are simply counts of single-cell spikes from 4918 or 3150 multiple superimposed sweeps, little or no indication of such distribution is ever seen in single traces or in a few superimposed traces.

Overall correlations (total sweep) of single-cell spike distributions with their respective evoked potential waveforms yielded significant correlation coefficients for 66 percent of the pairs. Correlation coefficients ranged from 0.14 to 0.88 with 58 percent significant at the .001 level, 6 percent at the .01 level, and 2 percent at the .05 level of probability.

Components of the evoked response in which there are deflections both negative and positive to the baseline are duplicated by a single-cell frequency distribution only if the spontaneous firing rate is greater than zero. The relation described here between the probability of firing of a single cell and the waveform of the evoked potential holds in general for responses of single cells to both visual and somatic stimuli recorded from a number of sensory and association cortical areas, for the cerebellar vermis, and for responses of single cells to two sensory stimuli separated by a short interval. That is, if a cell responds to more than one sensory stimulus, the probability of response to each stimulus is given by the respective waveform of the evoked potential.

We conclude from these data that no component (positive or negative, early or late) of the asynchronous evoked potential recorded in this way is uniquely related to responses of specific cell populations or to specific portions of cells, since from each cell a probability curve can be obtained which closely resembles the waveform of the entire evoked response. This outcome suggests, therefore, that whatever potential sources contribute to the evoked potential, they are directly related to or reflected in the firing of a single cell.

The data appear to support at least two interpretations of evoked potential electrogenesis: (i) the evoked potential may consist of summated and asynchronous discharges from cell bodies, with cells in a given localized area having approximately the same probability of firing, or (ii) the evoked potential may be compounded of electrical activity from a number of sources either local or distant, for example, apical, somatic, or basal dendritic postsynaptic potentials and local potentials associated with electronic spread from dendrites.

Although our records for both cells and waves are from microelectrodes deep in the cortex, some more specific conclusions can be drawn. It appears, for example, that either the recorded evoked potentials do not reflect dendritic activity at all and that both positive and negative waves recorded are local in origin, or that there is, in fact, an important relation between dendritic activity and the production of spikes from the cell body .

Whatever mechanisms actually underlie the observed correlation between probability of firing of single cells and the waveform of the evoked potential, it is certain that, contrary to conclusions from other studies (*4*), knowledge of the waveform of the evoked potential does, to a great extent, enable prediction of the response pattern of a particular cortical cell.

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Duplexity Theory of Taste

It has been unfortunate for the study of the neural bases and psychophysics of the chemical senses that the underlying physical continua are unknown; there is no wavelength or frequency for taste and smell. Consequently, most studies of taste, psychophysical and neural, are based on its four psychophysical "primaries," sweet, sour, bitter, and salty. These four primaries have never been too convincing, as is evidenced by von Békésy's recent psychophysical "duplexity" theory of taste (*1*), in which bitter and sweet are grouped together with warm as one factor, and salty and sour with cold as the other. These relationships were chosen on the basis of the interactions within each group and lack of interaction between groups.

Recently it has been shown that the neural basis of taste involves many fiber types rather than two or the classic four (*2*). Since von Békésy intends to refer his data to physiological mechanisms, perhaps the relation of these two sets of data should be discussed. The neural argument (*2*) concluded that the neural basis of taste is much like audition in that there are no "primaries," but that there are many fiber types ranging along the stimulus continuum.

Thus, an investigator choosing stimuli in either taste or audition, if he did not know the relevant stimulus continua, might choose two groups of stimuli with large intragroup interactions and small or nonexisting intergroup interactions. Clearly it would be improper to derive from this a two-factor theory for audition. The proper procedure in both modalities would involve either the use of more stimuli or the discovery of the continuum.

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