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Classical conditioning rapidly induces specific changes in frequency receptive fields of single neurons in secondary and ventral ectosylvian auditory cortical fields

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To determine if learning-induced changes in the response of auditory cortical neurons to a conditioned stimulus (CS) reflect general changes in cellular excitability or alterations in signal processing that are specific to that stimulus, we determined frequency receptive fields (FRFs) of single neurons in secondary and ventral ectosylvian auditory fields of the cat during classical conditioning. Associative changes in FRFs of most cells were specific to the frequency of the CS, established rapidly and reversed by extinction. Thus, learning causes specific changes in cortical processing of sounds whose significance is acquired.

Systematic changes in sensory cortical evoked responses to stimuli that signal reinforcement are among the most consistent and pronounced neurophysiological correlates of learning[2,3]. Studies of auditory cortex, which has been investigated most extensively over more than 30 years, have revealed that evoked potentials[4], multiple unit activity[5,15] and single-unit discharges[6,7] to an acoustic conditioned stimulus (CS) are altered by associative processes[8]. One hypothesis is that such physiological plasticity reflects general changes in cortical excitability[9,10]. An alternative is that associative learning specifically alters the processing of CS information in sensory cortex. To resolve this issue, we recorded single-unit activity in auditory cortex during a modified classical conditioning procedure. In addition to recording CS-evoked activity during training, we also determined the frequency receptive field (FRF) of each cell before and after acquisition of the pupillary dilation conditioned response[11]. If learning-induced discharge plasticity merely reflects a general change in excitability, then cells should increase or decrease their response across frequencies, without regard to the particular frequency of the CS. On the other hand, the processing specificity hypothesis predicts that changes in response should be specific to the frequency of the conditioned stimulus. Our findings support the latter hypothesis.

Adult cats (3–5 kg) were prepared chronically for single-unit recordings as reported previously[6]. A sequence of isointensity tones (range across sessions: 20–30 ascending frequencies, 0.1–24.0 kHz, 300 ms duration, 1500 ms intertone interval, repeated 10–15 ×, 30–80 dB) was presented under computer control via a calibrated delivery system to determine the FRF. The CS was a tone identical to one of the stimuli used to obtain the FRF. All tones were presented contralateral to the recording site. The unconditioned stimulus (US) was mild electrodermal stimulation to a forepaw, sufficient to cause a transient, nonhabituating pupil dilation. The conditioned response (CR) was pupillary dilation to the CS. FRFs were determined immediately before and after sensitization (CS/US unpaired, 20 trials), conditioning (CS/US paired, US presented 700–2000 ms after CS offset, 20–45 trials) and extinction (CS alone, 20 tri-

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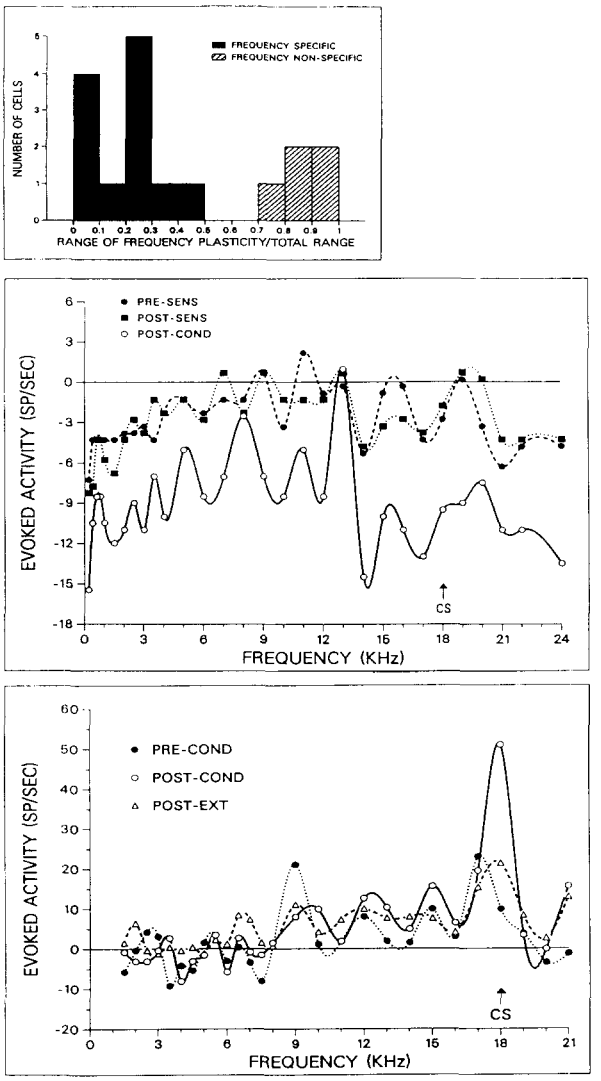

Fig. 1. Top: distribution of cells by index of frequency specificity. Middle: FRFs for a non-frequency-specific cell. Before training (pre-sens) it was inhibited at low frequencies (0.1–8 kHz) and at some high frequencies (e.g. 19–24 kHz); there was no significant change in this function immediately following sensitization (post-sens) during which the CS of 18 kHz was presented unpaired with the US. However, immediately after the conditioning phase, during which the animal acquired a pupillary dilation CR, the amount of inhibition increased significantly at almost every frequency. Although the amount of increase was somewhat greater around the CS frequency (i.e., 14–24 kHz), the range of frequency plasticity was as large as the range of preconditioning (inhibitory) responsivity (0.1–24 kHz); the specificity index of this cell was therefore 1.0. Bottom: FRF's for a frequency-specific cell. Prior to training, it was excited slightly at several frequencies above 8 kHz (pre-cond). Immediately after acquisition of the pupillary dilation CR to a CS of 18 kHz, the excitatory response was increased significantly only at the CS frequency (post-cond). Following behavioral extinction, the increased response at the CS frequency was abolished (post-ext).

als). Stimulus density was maintained constant at an average of 1 per 20 s. FRFs were compared by two-way ANOVA with replications[13]. Animals were maintained on gallamine (20 mg/h, i.v.) and artificial respiration to eliminate the confounding effects of stimulus inconstancy and proprioceptive feedback[14].

Data were obtained from 20 cells, one per training session, from the secondary (AII) and ventral ectosylvian (VE)[1] auditory cortical fields. Pupillary CRs developed rapidly during stimulus pairing (mean = 21.9 trials to statistical criterion, 14.6 min). Consistent with previous findings[6], almost all (19/20) cells rapidly developed discharge plasticity during conditioning (mean = 11.7 trials, 7.8 min). Most importantly, the FRFs of 80% of the neurons changed following conditioning (AII, n = 9/10; VE, n = 7/10). The effects were associative as FRFs were unchanged by sensitization procedures.

To quantify the degree of CS specificity of FRF plasticity, we calculated the ratio between two frequency range measures: the range of frequencies *above and below* the CS frequency to which the cell developed discharge plasticity divided by the total range of frequencies to which the cells responded. Cells whose specificity ratio was ≤ 0.5 were classified as frequency-specific. Of the cells, 75% (12/16; Fig. 1 top) met this criterion (AII, 5/9; VE, 7/7). An example of a cell exhibiting a non-specific associative FRF change is presented in Fig. 1 (middle). The postconditioning FRF indicates an enhancement of inhibition to most frequencies. An example of a cell that developed a CS-specific change in its FRF is shown in Fig. 1, bottom. This neuron developed a significant increase only to the frequency of the CS (18 kHz). The conditioning effect was reversed by extinction (Fig. 1, bottom). This reversal effect occurred for all cells tested for extinction (specific, n = 7; non-specific, n = 3).

These findings raise two immediate questions: (i) what variables govern the development of CS specific associative changes in FRFs; (ii) are extinction effects caused by the omission of the US or simply due to dissipation of the plasticity with time? Fig. 2 presents data that address both issues. This cell was studied during sequential training using two different frequencies as the CS, and after extinction. Initially, there was no significant change in the FRF following conditioning using a CS of 12 kHz (Fig. 2, top). A

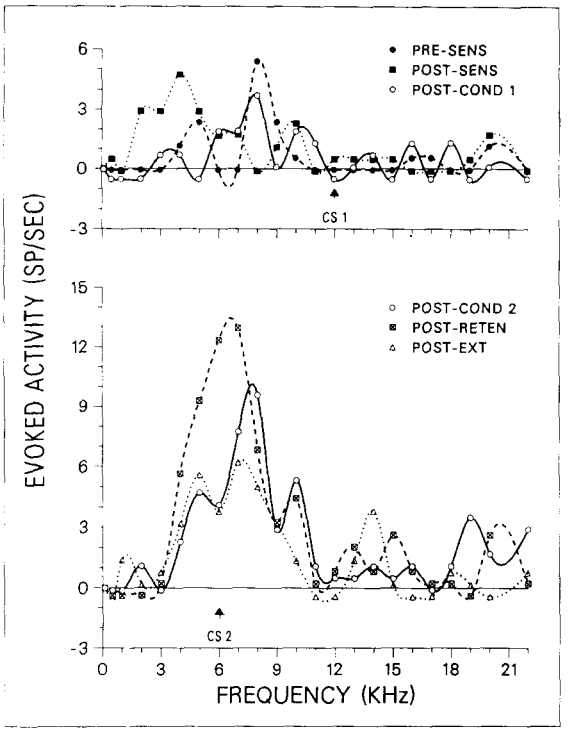

Fig. 2. Top: FRFs for a frequency-specific cell recorded during training with two frequencies. Prior to training, the largest response was an excitation at 8 kHz (pre-sens); a non-associative sensitization effect occurred as excitation at 2–4 kHz (post-sens), but this disappeared after acquisition of the pupillary dilation CR (post-cond 1). Bottom: when conditioning was then run with a CS of 6.0 kHz, the response increased significantly at 5–10 kHz (compare post-cond 2 with post-cond 1). Following a 20-min period of silence, the increased responses near the CS frequency not only remained, but were further increased, with the largest responses at 5–6 kHz (post-reten). Immediately after extinction, the frequency-specific increases were diminished (post-ext).

second conditioning phase was then run using a CS of 6 kHz (Fig. 2, bottom). This frequency was selected because it was closer to the region of the small excitatory responses evident at the start of the session (Fig. 2, top). In contrast to the lack of plasticity in the first postconditioning FRF, the second postconditioning FRF (CS = 6 kHz) revealed a significant increase in the responses to 5–10 kHz. Such data were obtained in 3/3 neurons. These findings suggest that the development of FRF plasticity may require at least a small initial response to the CS, rather than arising de novo.

To determine if extinction effects are due to the dissipation of conditioning effects with time or to the removal of the US, FRF of 4 cells were obtained after

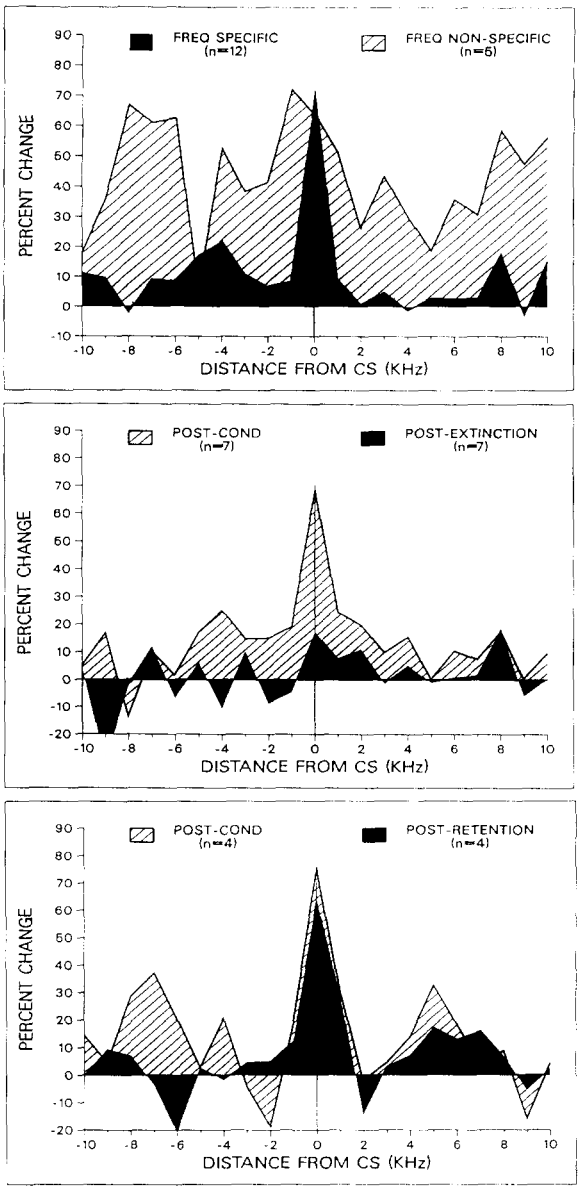

Fig. 3. Changes in discharge response as a function of distance (in kHz) from the frequency used as the CS. Top: the largest change following conditioning was at the CS frequency for the frequency-specific cells; large changes were found for non-frequency-specific cells without limitation to the region of the CS. Middle: extinction data for frequency-specific neurons shows the reversal of the associative response changes after presentation of the CS alone with concommitant behavioral extinction. Bottom: retention data indicate that the CS-specific effect of learning does not dissipate spontaneously.

a period of silence equal to the maximum time required to obtain behavioral extinction. Using this test of retention of FRF plasticity, learning-induced effects did not dissipate (2/3 frequency-specific, 1/1 fre-

quency-non-specific). In fact, for the cell in the current example, the effect was greater at this 'post-retention' FRF than the postconditioning FRF, (Fig. 2, bottom). When extinction was then run, the conditioning effects diminished greatly ('post-ext').

To determine the degree of specificity relative to the CS frequency, postconditioning data from each cell were normalized as a positive percent change from the presensitization FRF and expressed as the distance in kHz from the CS. This analysis revealed that the greatest change for the frequency-specific neurons was to the CS frequency (71%) while the change in response to all other frequencies was less than 25% of control values (Fig. 3, top). This exceptional degree of specificity is noteworthy. In contrast to the frequency-specific neurons, those classified as frequency-non-specific developed a 30–72% change to most frequencies, with no distinction between the magnitude of change to the CS and stimuli as distant as 10 kHz from the CS.

This normalization analysis also revealed that the reversal of the conditioning effects with extinction resulted from the omission of the US, not to the passage of time: extinction eliminated the frequency-specific plasticity (Fig. 3, middle) whereas the mere passage of time did not appreciably diminish the conditioning effects (Fig. 3, bottom).

These findings demonstrate that the frequency-receptive field of single neurons in the secondary and ventral ectosylvian auditory cortical fields can be changed rapidly by associative processes and that such changes may be stable unless stimulus significance is reversed by extinction. Because these effects were mainly specific to the CS, processing specificity is a feature of learning-induced cortical function. Thus, the frequency-receptive field in auditory cortex is a dynamic characteristic that can reflect the acquired significance of acoustic stimuli. Although the relevant neural mechanisms remain to be studied, processing specificity is probably not projected to the cortex from the thalamus because the frequency organized subdivision of the medial geniculate nucleus is non-plastic during learning[12]. The processing specificity may reflect cortical mechanisms involved in selective attention and discrimination. Further study is necessary to determine the time course of the retention of learning-induced receptive field plasticity, as well as to characterize receptive field plasticity for other stimulus parameters, cortical auditory fields, and sensory systems.

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1 Diamond, D.M., *Physiological Plasticity of Single Neurons in Auditory Cortex of the Cat During Learning*, Doctoral Dissertation, University of California, Irvine, 1985 (unpublished).
2 Diamond, D.M. and Weinberger, N.M., Physiological plasticity of single neurons in auditory cortex of the cat during acquisition of the pupillary conditioned response II, secondary field (AII), *Behav. Neurosci.*, 98 (1984) 189–210.
3 Galambos, R., Sheatz, G. and Vernier, V., Electrophysiological correlates of a conditioned response in cats, *Science*, 123 (1956) 376–377.
4 Hall, R.D. and Mark, R.G., Fear and modification of acoustically evoked potentials during conditioning, *J. Neurophysiol.*, 30 (1967) 893–910.
5 John, E.R., Higher nervous functions: brain functions and learning, *Annu. Rev. Physiol.*, 23 (1961) 451–484.
6 Kitzes, L.M., Farley, G.R. and Starr, A., Modulation of auditory cortex unit activity during the performance of a conditioned response, *Exp. Neurol.*, 62 (1978) 678–697.
7 Marsh, J.T., Worden, F.G. and Hicks, L., Some effects of room acoustics on evoked auditory potentials, *Science*, 137 (1962) 280–282.
8 Miller, R.G., Jr. *Simultaneous Statistical Inference*, Springer, New York, 1981.
9 Morrell, F., Electrophysiological contributions to the neural basis of learning, *Physiol. Rev.*, 41 (1961) 443–494.
10 Olds, J., Disterhoft, J.F., Siegel, M., Kornblith, C. and Hirsh, R., Learning centers of rat brain mapped by measuring latencies of conditioned unit responses, *J. Neurophysiol.*, 35 (1972) 202–219.
11 Oleson, T.D., Ashe, J.D. and Weinberger, N.M., Modification of auditory and somatosensory system activity during pupillary conditioning in the paralyzed cat, *J. Neurophysiol.*, 38 (1975) 1114–1139.
12 Ryugo, D.K. and Weinberger, N.M., Differential plasticity of morphologically distinct populations in the medial geniculate body of the cat during classical conditioning, *Behav. Biol.*, 22 (1978) 275–301.
13 Weinberger, N.M., Hopkins, W. and Diamond, D.M., Physiological plasticity of single neurons in auditory cortex of the cat during acquisition of the pupillary conditioned response; I, primary field (A1), *Behav. Neurosci.*, 98 (1984) 171–188.
14 Weinberger, N.M., Diamond, D.M. and McKenna, T.M., Initial events in conditioning: plasticity in the pupillomotor and auditory systems. In G. Lynch, J.L. McGaugh, and N.M. Weinberger (Eds.), *Neurobiology of Learning and Memory*, Guilford, New York, 1984, pp. 197–227.
15 Weinberger, N.M., Diamond, D.M. and McKenna, T.M., Plasticity of frequency tuning of single auditory neurons in auditory cortex during learning, *Soc. Neurosci. Abstr.*, 10 (1984) 245.