Role of Context in the Expression of Learning-Induced Plasticity of Single Neurons in Auditory Cortex

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Classical conditioning produces frequency-specific plasticity of receptive fields (RFs) of single neurons in cat auditory cortex (Diamond & Weinberger, 1986). In this article we show that although plasticity may be observed during both training trials and determination of RFs, it is usually expressed in a qualitatively different form (e.g., decreased response during conditioning vs. increased response to this same conditioned stimulus in the postconditioning RF). This differential expression of learning-induced plasticity provides evidence for a role of context in neurophysiological mechanisms of learning in auditory cortex. A model of cortical neurons functioning within a mosaic of influences is presented. The Functional Mosaic model views the induction and expression of plasticity as separate processes.

In this article we address the role of context in the expression of learning-induced neuronal plasticity. For present purposes, we consider context to refer to the different circumstances under which the same stimulus may be presented. Although the role of context in learning has been studied extensively at the behavioral level in both human and animal studies (see Mackintosh, 1985, for a review), this issue has received attention in studies of the neurobiology of behavior primarily only in the last decade. Specifically, the effects of brain stimulation, lesions, and neurotransmitter depletion on the capacity of animals to use information about contextual cues have become well documented (Archer, Mohammed, & Jarbe, 1983; Kesner & Hardy, 1983; Sara, 1985; Winocur & Gilbert, 1984). In addition, studies of physiological activity recorded in animals during the performance of learned tasks have identified contextual variables as a major component of learning-induced neuronal activity (Brown, 1982; Kubic & Ranck, 1983, 1984; Manetto & Lidsky, 1983; Wible, Findling, Shapiro, Lang, & Olton, 1986).

The latter studies have revealed that during the performance of learned tasks, neurons respond to the same stimuli differently depending on the circumstances in which environmental stimulation is presented.

In the present study, we have characterized the role of context in the expression of learning-induced neuronal plasticity. The finding was actually serendipitous because the explicit purpose of the experiment did not include an investigation of the role of context in discharge plasticity. In order to appropriately introduce the rationale that led to the experimental paradigm, it is necessary first to provide background information on the purpose of this experiment.

There is extensive documentation indicating that associative learning alters the responses of sensory systems to stimuli when they acquire behavioral significance (for reviews, see John, 1961; Sokolov, 1977; Thompson, Patterson, & Teyler, 1972; Weinberger & Diamond, 1987). The effects of associative processes on stimulus-evoked activity are prominent at the highest levels of sensory systems, with the most consistent evidence of plasticity in sensory cortex (e.g., olfactory: Freeman & Skarda, 1985; somatosensory: Oleson, Ashe, & Weinberger, 1975; Voronin, Gerstein, Kudryashov, & Joffe, 1975; visual: Shinkman, Bruce, & Pfingst, 1974; Morrell, Hoeppner, & deToledo-Morrell, 1983). In studies of auditory cortex, which is the most thoroughly investigated sensory cortical region, learning-induced changes in evoked activity have been documented extensively over the last 3 decades (for a review, see Weinberger & Diamond, 1987).

Although associatively induced plasticity in sensory systems is well documented, a fundamental issue regarding the nature of the changes was resolved only recently. It was not known whether such plasticity actually reflects changes in information processing that are specific to the conditioned stimulus (CS) or whether they merely reflect nonspecific changes in neuronal excitability. Historically, this issue has received some attention, with experimenters concluding that learning-induced changes in sensory systems are largely if not completely attributable to nonspecific changes in arousal level.
(e.g., Hall & Mark, 1967; Kitzes, Farley, & Starr, 1978; Miller, Pingst, & Ryan, 1982). Elsewhere we have presented an analysis of these experiments that explains why the conclusion of nonspecificity is unsupported by the data (Weinberger & Diamond, 1987, 1988).

The explicit purpose of the present experiment was to test two mutually exclusive hypotheses derived from the nonspecific and from the processing specificity viewpoints. If learning produces a nonspecific change in the responsivity of auditory cortex, then it should produce a broad generalization gradient of altered auditory sensitivity; effects should not be more pronounced for the frequency of the CS than for other tones. In contrast, if learning-induced plasticity reflects a specific change in the processing of the CS, then it would be revealed as a narrow band change in response that is significantly greater for the frequency of the CS than for other acoustic stimuli.

In order to resolve the issue of the specificity of learning-induced changes, we recorded discharges from single neurons in auditory cortex during a modified classical conditioning procedure. In addition to recording CS-evoked activity during training trials, we also determined the frequency receptive field (FRF) of each cell before and after acquisition of a behavioral conditioned response (CR): pupillary dilation. We found a "narrow bandwidth" change in evoked activity that was centered on the CS frequency (Diamond & Weinberger, 1986). The specific changes developed rapidly, in less than 30-40 conditioning trials (within 25 min), were reversed just as rapidly during behavioral extinction, and were maintained in the absence of extinction training. The finding that cells exhibited CS-specific changes with learning supported the processing specificity hypothesis. (In the Appendix we discuss these findings from the standpoint of auditory cortical neurons function as adaptive filters.)

We designed the paradigm of the current experiment as a test of the issue of specific versus nonspecific changes in CS-evoked activity with learning. The issue of context became manifest by the nature of the findings. We assumed that the changes in evoked activity that developed to the CS during training would be similar to the changes in response to that same frequency during the determination of the FRF after training. However, the characteristics of plasticity to the CS that were expressed in the posttraining FRF were usually different from the plasticity that developed to the CS during training. In these cases, the effect was not simply a matter of degree, but was actually a difference in the direction of change in evoked activity. For example, a neuron that developed a decrease in evoked activity to the CS during conditioning then exhibited an increase in evoked activity to the CS in the postconditioning FRF. We provide evidence that this difference in evoked response can be attributed to neither nonassociative factors nor to an inconstancy of stimulus detection at the periphery. Rather, the effects resulted from associative processes intrinsic to the nervous system.

The difference in the expression of plasticity to an identical stimulus presented in two different circumstances argues strongly for a role of context in neurophysiological mechanisms of learning. In this article we present data relevant to context, including details about frequency specificity where necessary. We also present a model called the Functional Mosaic that is based on the concept that cortical cells are components of different functional systems, depending on the context of stimulation.

Method

Surgical Preparation

Details of the preparation are identical to those published previously (Diamond & Weinberger, 1984). Briefly, under general anesthesia (sodium pentobarbital, 35 mg/kg), adult cats (n = 13; 2.5-6.0 kg) had threaded metal cylinders embedded in a pedestal of dental acrylic fixed to the calvarium. Body temperature was maintained by a thermostatically controlled warm water pad during the surgery and recording sessions. Ophthalmic ointment (Terramycin) was applied to prevent corneal drying. Antibiotics (Panalog and Delagon) were applied locally to exposed skin surfaces, and Bicillin (300,000 U, im) was administered for 2 days following the surgery. The subjects recovered in an incubator, and training began 1-2 weeks later.

General Procedure

At the beginning of a training session, the animals underwent neuromuscular blockade induced by gallamine triethiodide (Flaxedil, 10 mg/kg, ip). Under laryngoscopic control, the trachea was intubated with a pediatric catheter coated with a local anesthetic (Xylocaine). The animal was then artificially ventilated with a Harvard respirator and positioned in a frame to which the pedestal was attached. All procedures were carried out with the animal located in an acoustically isolated chamber (IAC 1202). Neuromuscular blockade was maintained with a constant intravenous infusion of Flaxedil (20 mg/hr). Expired carbon dioxide levels were monitored with a Beckman Medical Gas Analyzer LB2 and maintained at approximately 3.5%.

Acoustic stimulation was delivered to the ear contralateral to the recording site by a Beyer DT48 speaker. The earphone was attached to an earpiece that was inserted into the external auditory meatus. Prior to each recording session, the output of the earpiece was calibrated in situ using a probe tube inserted through a sealable port in the sound delivery tube. Sound pressure level was calibrated using a calibrated 1/2-in. (1.27 cm) condenser microphone, a Bruel and Kjaer sound level meter, and a Hewlett-Packard Wave Analyzer and then stored as a table of frequency versus maximum intensity in an LSI/11 computer. Throughout each session, acoustic stimuli were presented under computer control (rise-fall 5 ms, Coulbourne audio gate). Sound frequency was set under computer control via a voltage-controlled gate of a Wavetek oscillator (0.1-24 kHz), and intensity was set with a Coulbourne computer-controlled attenuator (range = 0-128 dB). Stimulus intensities are expressed as decibels above a reference of 20 μN/m2. The acoustic CSs were tones (range = 100-300 ms [constant within a recording session]), 0.7-24 kHz, 30-85 dB). The unconditioned stimulus (US) was electrodermal stimulation consisting of a 200-ms train of 50-Hz pulses (5 ms each) produced by a Grass S-44 constant-current stimulator via a stimulus isolation unit. Stimulus intensity (2-9 mA) was set at the beginning of each session to produce a brief (2-5 s) pupillary dilation. The US was delivered to the forepaw contralateral to the recording site.

Single-unit discharges were recorded with tungsten electrodes (1 μm tip diameter, 5-25 μm shaft diameter) insulated with Epoxylite (1-3 MΩ impedance at 1 kHz), with cortical access via a small burr hole. A Narashige microdrive permitted movement of the electrode in 1-μm steps. Cellular discharges were amplified by a Dagan 2400 preamplifier (0.3-3 kHz band-pass, 10,000 x gain), the output of
which was displayed on a storage oscilloscope, recorded on a direct channel of a Hewlett-Packard 3964A tape recorder and passed through a voltage detector that produced a single pulse for each discharge exceeding a selected level. The pulses were led to an LSI/03 computer for analysis. A single unit was identified by visual inspection of the waveform. Recording was discontinued if the waveform became unstable or if it could not be unambiguously separated from other waveforms. The stringent requirement of obtaining data throughout all training and testing procedures from only one neuron resulted in complete loss of data if a recording was discontinued before postconditioning data were obtained. Approximately half of the data sought were eliminated for this reason.

Pupillary size was monitored by an infrared pupillometer (Casaday, Farley, Weinberger, & Kitzes, 1982) positioned in front of the eye contralateral to the recording site. Care was taken to avoid possible discomfort caused by drying of the corneas by covering them with ophthalmic ointment. The output of the pupillometer was amplified by a DC amplifier and written out on a Grass Model 7 polygraph.

**Experimental Design**

**Rationale and Overview of Experimental Procedures**

The explicit goal of this study was to investigate the effects of learning on the FRF properties of single auditory cortical neurons. This was achieved by recording discharges evoked by presenting a sequence of isointensity tones at predetermined times during the course of a training session. The evoked activity to the frequencies constituted an FRF. The particular frequency of the CS during classical conditioning periods of each session was one of the frequencies used in the determination of the FRF.

A general overview of the experimental protocol is as follows: Following isolation of discharges from a single unit, an FRF was obtained. After 20 min of silence, a second FRF was obtained in order to test the stability of the frequency response properties. Immediately following the retest, classical conditioning procedures were initiated. An FRF was obtained in conjunction with each phase of behavioral training: sensitization, conditioning, extinction, and retention. One cell was studied in each recording session. A summary of the protocol is given in Figure 1. The following section provides more detailed accounts of the training procedures and determinations of the FRF.

**Experimental Protocol**

An FRF was obtained in the following manner: A sequence of tones, composed of ascending frequencies (range = 0.1–24 kHz) was presented to the animal. The intensity was constant at each frequency, with a range of 30–80 dB across cells. For each cell, the intensity was set at a level that was at least 10 dB above threshold, but not so great that a saturation of an evoked response at any intensity was evident. Each sequence was composed of a constant number of tones (range = 16–30 tones across cells) of constant duration (range = 200–900 ms across cells), with a constant intertone interval (range = 1,000–2,000 ms across cells). The duration of each sequence of tones ranged from 40 to 60 s across cells. After an intersequence interval of 15–30 s, the sequence of tone presentation was repeated. The number of repetitions was constant for each cell (range = 9–15 across cells). The time required to obtain one FRF was 10–15 min.

After 20 min of silence, a second FRF was obtained to determine FRF stability prior to training. These pretraining functions are referred to as Presensitization 1 and Presensitization 2, respectively. Data reported in this article are restricted to neurons in which these two FRFs were not significantly different.

Immediately after the second FRF determination, the sensitization phase was initiated. The frequency of the CS was selected after review of the two FRFs. The CS was never the frequency to which the cell responded maximally (best frequency) in order to avoid a possible ceiling effect. In the case of cells that responded to a narrow range of frequencies, the CS was selected at the low or high range of responsivity. In the case of cells that were broadly tuned, the CS was a frequency that evoked a weak response.

The frequency, intensity, and duration of the CS presented in sensitization, conditioning, and extinction were identical to one of the tones within the sequence of tones used to obtain the FRF (except for one training session in which three contiguous frequencies in the sequence were used as the CS). During sensitization, 15 CSs and 15 USs were presented in an explicitly unpaired fashion, at pseudo-random intervals at an average density of 2 per 40 s, with the restriction that stimuli not occur within 10 s of each other (range = 10–30 s/stimulus within a session). The sensitization phase served as a control for nonassociative factors. After these sensitization trials were presented, an FRF was obtained once again. This is referred to as the post-sensitization FRF.

After determining the postsensitization FRF, five more trials of each sensitization trial type (CS–US) were given for the following reason: The FRF determination itself might have had a nonassociative effect on the behavioral response, the neural response, or both to the CS. Extending the sensitization phase for five additional trials would have revealed such effects. However, this precautionary mea-
sure turned out to be unnecessary because obtaining the FRF induced no obvious changes in either neural or behavioral measures. Following the last trial of sensitization, conditioning began without delay. During conditioning, the CS and US were paired on every trial, with the US presented at a fixed interval (trace period) after the CS offset. The trace period ranged from 700 to 3,000 ms for different cells. Stimulus density was maintained at 2 per 40 s, and the average intertrial interval was 40 s (range = 20–60 s within a session). Generally, 30 conditioning trials were given. After the last conditioning trial, a postconditioning FRF was obtained immediately.

In three sessions, an additional conditioning phase was run. This phase involved the selection of a frequency of a second CS that was different from that of the CS used during the initial conditioning phase. Although the CS frequency was different, the stimuli used to obtain the FRF were constant throughout the duration of a training session. In these recording sessions, an extinction phase followed each of the conditioning phases (see the Extinction section).

Frequently, the unit waveform remained stable after conditioning. When this occurred, (a) extinction or (b) retention procedures were initiated to further study response properties.

**Extinction.** Following the determination of the postconditioning FRF, an extinction procedure was run in seven cases. Extinction consisted of the presentation of the CS alone. Stimuli were presented at the same rate as in the sensitization phase (2 per 40 s). Generally, the magnitude and duration of the CR diminished within 5–40 trials. The maximum time required for behavioral extinction was 20 min. After the extinction phase was completed, the FRF was obtained again; this FRF is referred to as postextinction.

**Retention procedures.** Following the determination of the postconditioning FRF, a retention phase was run in four cases. The retention phase was simply a period of 20 min without the presentation of any stimuli. After this period of silence, the FRF was determined once again. This FRF is called postretention. The rationale for this procedure is as follows: First, it may be argued that the effects of the extinction procedure occur because of the passage of time per se rather than because of the unreinforced presentation of the CS. Retention data taken at the same interval as used to obtain postextinction FRF data would resolve this issue. Second, the retention data would provide an initial characterization of the degree of durability of associative effects produced by the conditioning phase.

**Data Analysis**

**Analysis of Training Trials**

The effects of training on pupillary behavior were assessed as described previously (Diamond & Weinberger, 1984; Weinberger, Hopkins, & Diamond, 1984). The pupillometer write-out was measured immediately preceding the presentation of the CS throughout training and preceding the US for sensitization trials (baseline measure). The peak amplitude of dilations to those stimuli was measured and the baseline level was subtracted, which yielded a difference score hereafter referred to as the pupillary response. The number of trials to criterion for pupillary conditioning was defined as five consecutive trials during the conditioning phase, all of which had pupillary responses greater than the average of the last five trials of the sensitization phase. The probability of this occurring by chance is < .03 (sign test; Siegel, 1956).

Spike data were stored in an LSI 11/03 computer during the 1.5 s immediately preceding a trial, during the trial, and for 3 s after the offset of the last stimulus of the trial (CS or US during sensitization, after the US during conditioning, and after the CS during extinction). The time of occurrence of each discharge (1-ms resolution) was recorded and later compiled into histograms with bin widths ranging from 1 to 50 ms. The average rate of activity per second was determined for every pretial period and every period of tone-evoked activity under analysis. The pretial period average was subtracted from the evoked activity for each trial, yielding a difference score. The pretial scores are hereafter referred to as background activity and the difference scores as evoked activity. The number of trials to criterion of discharge plasticity was defined as five consecutive trials during the conditioning phase, all of which had to be greater or smaller values than the average of the last five trials of the sensitization phase (p < .05).

**Analysis of Frequency Response Functions**

The procedure for analysis of evoked activity was as follows: The histogram of the FRF was visually inspected for evidence of prominent evoked activity. Once a component was identified, a temporal window was set within its confines. The response to every frequency in the sequence was then analyzed using the same window (e.g., for each of 30 frequencies, discharge activity occurring from 30 to 100 ms after tone onset was quantified). The same window was applied to every frequency response function determined throughout the experiment. For every FRF, there was a 1.5-s presequence period that served as a measure of background activity. The background activity was then subtracted from the evoked activity. This was done for two reasons: First, this technique eliminates apparent changes in evoked activity that reflect a general change in excitability of a cell rather than a specific effect that occurred during acoustic stimulation. Second, it provided for the quantification of stimulus-evoked activity that was less than ongoing background activity (i.e., inhibition). All data were normalized to spikes per second.

**Statistical Analysis of the Frequency Response Function**

The first two FRFs served as a test of the stability prior to training. In order to assess the degree of similarity of the two FRFs, their Pearson correlation coefficient was determined. A statistically significant correlation coefficient (p < .05) was the criterion for classifying the cells as exhibiting a stable FRF prior to training.

In order to test for changes in FRFs across training phases, the data were compared using an analysis of variance (ANOVA) with replications. This procedure provided for the identification of significant changes in the responses to particular frequencies in the postsensitization and postconditioning FRFs, and if applicable, postextinction and retention FRFs.

**Determination of Recording Site Location**

At the conclusion of each training session, a small electrolytic lesion was produced by passing anodal current through the recording electrode. After the final session, the animal was given an overdose of Nembutal and perfused either through the carotid arteries or heart with 0.9% saline followed by 10% formalin; the brain was then removed and stored in formalin. Frozen serial sections (50 μm) were taken throughout auditory cortex and stained with cresyl violet.

**Results**

**Analysis of Pupillary Behavior**

**Behavior During Training Trials**

Pupillary dilation responses were recorded during all sessions. At the beginning of sensitization, the acoustic stimulus generally elicited a brief dilation. By the end of the sensiti-
zation phase, dilation responses were reduced in both amplitude and duration. The US produced consistently large dilations throughout the session. During the conditioning phase, responses evoked by the CS increased rapidly over the trials, as repeatedly demonstrated in previous experiments (e.g., Diamond & Weinberger, 1984; Weinberger, 1980; Weinberger et al., 1984). The average CRs during Conditioning Trials 6-10 typically exceeded the largest acoustically elicited responses of the preceding sensitization phase. The magnitude and duration of the CR attained asymptote by Trials 20-25 and maintained high values thereafter (see Figure 2). The pupillary CR attained statistical criterion in 17 of 20 sessions ($M = 21.9$, $SD = 15.2$, range = 6–50 trials). In 9 sessions, unit recordings were maintained during an extinction phase that followed conditioning. During extinction, CS-evoked dilations generally decreased, returning to sensitization levels within 5–40 trials. A typical example of pupillary behavior during a single session is presented in Figure 3.

**Behavior During the Determination of FRFs**

As described in detail in the Method section, FRFs were determined by presenting multiple (10–15) sequences of isointensity tones. The tone used as the CS during training trials was identical in all physical parameters (e.g., frequency, intensity, and duration) to one of the stimuli used to obtain the FRF. The only difference in the presentation of this tone between training and FRF determination was contextual (i.e., for the former, the CS served as a predictor of the US, and for the latter, the CS was embedded in a sequence of tones). The US was never presented during the determination of the FRF. Therefore, it was possible to compare responses with the CS when it was presented under the two circumstances. Differences in pupillary and neuronal responses to the CS between conditioning and FRF determination were interpreted to reflect the effects of different contexts.

Behaviorally, animals exhibited stereotyped responses to the sequences of tones used to obtain the FRF: The baseline or tonic level of dilation remained relatively stable or decreased (indicating pupillary constriction), with small dilations interspersed throughout the sequence. In some cases, the subjects dilated to the initial tones with diminished responsiveness to later tones. The CS frequency was never in the initial part of the sequence in order to distinguish between selective responses to the CS and initial orienting responses. Examples of pupillary behavior during FRF determination, contrasted with behavior during conditioning, are presented in Figure 4. In no case did an animal selectively dilate to the CS frequency (or similar frequencies) presented during determination of an FRF.

**Analysis of Neuronal Activity**

**General Observations of Neuronal Activity in Secondary Auditory Cortex (AII) and Ventral Ectosylvian Auditory Field (VE)**

Unit activity in AII exhibited excitatory responses, inhibitory responses, or both across a wide range of frequencies (usually greater than 15 kHz), even at threshold intensity. Neurons that responded to tones had an evoked response latency range of 30–200 ms from tone onset, and tone offset, or both.

Neurons located in the region that has been considered posterior AII, along the anterior bank of the posterior ectosylvian sulcus (Schreiner & Cynader, 1984), differed in their acoustic response properties from those located more anteriorly (see Figure 5). In this and previous accounts (Diamond & Weinberger, 1986; Weinberger & Diamond, 1987), we refer to posterior AII as the VE. Units in VE were responsive to a restricted range of frequencies, and a best-frequency range could usually be determined. VE and primary auditory cortex (AI) neurons were distinguished on the basis of their range of frequency tuning and tone onset latency. AI neurons were more narrowly tuned and responded to sound at short onset latencies (less than 25 ms; Reale & Imig, 1980). VE neurons, on the other hand, responded to a broader range of frequencies than those in AI at comparable intensities. In addition, their latency to evoked response was relatively long (30–50 ms).

Although we did not attempt to obtain complete mapping data of VE, we consistently encountered a gradient of best-frequency tuning; high-frequency tuning predominated in dorsal VE and lower frequency tuning was ventral. VE is not likely to be an anterior extension of the tonotopically arranged posterior auditory field (PAF) described by Reale and Imig (1980) because the frequency organizations of VE and PAF are in opposite directions.

**Neuronal Activity During Conditioning Trials**

Single-unit data were obtained from 20 neurons in the AII–VE region. Although cells in AII and VE differed in their range of frequency tuning, their likelihood to develop associatively induced changes in discharge activity during training did not differ significantly. Six of 7 neurons in VE were plastic in both background and evoked activity, and 10 of 13 neurons in AII were similarly plastic. Additionally, there was no significant difference in the direction or rate at which plasticity developed during individual conditioning trials (all tests were Mann-Whitney and chi-square, $p > .1$).

Nineteen of the 20 neurons reached criterion for plasticity of evoked activity during conditioning. Changes in acoustic-evoked activity developed rapidly, reaching criterion in an average of 11.7 trials ($SD = 7.7$). Eight neurons developed an increase in response to the CS ($M = 10.0$ trials, $SD = 6.1$, range = 6–24), and 11 developed response decreases ($M = 13.0$, $SD = 8.8$, range = 6–35). The rates of development of increases and decreases in evoked activity did not differ significantly (Mann-Whitney, $p > .1$).

In 17 recording sessions, both pupil learning and plasticity of evoked activity occurred. In these sessions, we were able to determine whether there was a tendency for one measure to develop plasticity in fewer training trials than the other measure. In 9 of 10 sessions (binomial test, $p < .05$), decreases in evoked activity developed in significantly fewer training trials than did behavioral learning (pupil mean = 26.4 trials, neuronal mean = 12.2 trials, Wilcoxin matched-
Figure 2. Group pupillary learning curve. (The group pupillary learning curve for the 17 sessions in which pupillary dilation conditioned responses developed is shown. Data are expressed as the mean percentage of the maximum pupil dilation to the conditioned stimulus in each training session. SENS = sensitization and COND = conditioning.)

PUPILLARY BEHAVIOR
(n=17)

% OF MAXIMUM RESPONSE

15 20 25 30 35 40 45 50 55 60 65 70 75

1 2 3 4 5 6 7 8 9 10 11 12

SENS COND

BLOCKS OF 5 TRIALS

Figure 3. Pupillary behavior during training. (Pupillary behavior of 1 subject during training is illustrated. Filled triangles indicate the onset of the conditioned stimulus [CS], and the open triangles indicate the onset of the unconditioned stimulus [US]. The US [250 ms] was presented 700 ms after the offset of the CS [300-ms duration]. Notice that during sensitization trials [CS5, CS10], the tone evoked a low-level dilation, whereas the US [US5, US10] consistently evoked a large dilation. During conditioning, the evoked response to the CS was augmented by the fourth trial [CS4] and continued to increase in magnitude later in conditioning [CS12, CS40]. A decrease in the magnitude of the conditioned response was evident in the fourth trial of extinction [E4], and the behavioral response was virtually abolished later in extinction [E24].)

PUPILLARY BEHAVIOR

SENSITIZATION

C5 U5

C10 U10

CONDITIONING

CS3 CS12

CS4 CS40

EXTINCTION

E2 E10

E4 E24

pairs signed-ranks, \( p < .01 \)). Although there was a tendency for the rate of increases in evoked activity to develop earlier than the pupil, the difference was not significant (pupil mean = 17.0, neuronal mean = 11.3 trials, Wilcoxin test, \( p < .1 \)).

Analysis of FRFs

Stability Testing

The stability of the FRF was determined for each cell before each training session. After isolating the discharges of one neuron, the FRF was determined twice, separated by a 20-min period of silence. This test of stability took place just prior to the initiation of the sensitization phase. In 19 of 20 cases, the FRFs were significantly correlated (Pearson cor-
Figure 4. Contrast between pupillary behavior during training and frequency receptive field (FRF) determination. (Pupillary records were taken directly from polygraph records during four different recording sessions [numbered 1-4]. Pupillary dilation is up, constriction is down. In the left column are representative examples of pupillary conditioned responses to the conditioned stimulus [CS; duration indicated as a block above the line], preceding the unconditioned response to the unconditioned stimulus [US; duration indicated by the block below the line]. In the lowest record [Number 4, bottom left], the conditioned response attains the maximum level of dilation so that the unconditioned response to the US is not discernible. In the right column are pupil records obtained during one of the sequences of tones that was used to obtain the postconditioning FRF. As many as 15 of these sequences [range = 9-15 across sessions] were used to determine the FRF. Only one of the stimuli in the sequence of tones was identical to the CS used during training. For each subject [1-4], the CS within the sequence of tones is indicated by the arrow. Most important, the subjects did not respond to the CS within the sequence of tones as they had during training.)
Examples of FRF stability are presented in Figures 6 and 7. Cell T16D (see the top of Figure 6), located in VE, was the most responsive to frequencies in the range of 1.5-6 kHz. The most notable characteristic is the highly reproducible pattern of the FRF. For example, the two primary peaks at 2.5 and 4 kHz, with additional regions of responsiveness to frequencies above 6 kHz (e.g., 7-10, 15, and 17-22 kHz), are almost entirely overlapping between the two FRF tests. The correlation coefficient for this cell was .96 ($p < .001$). Another example of FRF stability in VE is provided in the lower part of Figure 6. This cell responded to stimuli from 4 to 10.5 kHz. The high degree of reproducibility of the FRFs is indicated by a significant correlation coefficient ($r = .65, p < .001$).

As mentioned previously, cells in AII were responsive to a broader range of frequencies than were those in VE. Examples of such broad tuning, which was highly stable, are in dashed lines. Dorso is up, anterior is left. AAF = anterior auditory field, AII = secondary auditory cortex, AI = primary auditory cortex, PAF = posterior auditory field, VPAF = ventral posterior auditory field, and VE = ventral ectosylvian auditory field.)

relation coefficient, $p < .05$), indicating that the FRF did not change spontaneously over the time domains used in this study.

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Physiological Plasticity and Context

The preceding results constitute a prerequisite for determining the expression of associatively induced plasticity during conditioning. In the following sections, we describe learning-induced changes in evoked activity to the CS when that stimulus was presented under two different circumstances: (a) during classical conditioning trials and (b) during the determination of the FRF. A summary of the effects of conditioning on the responses of each cell during training and determination of its FRF is presented in Table 1. For the majority of the plastic cells (13 of 19, 68%), the expression of plasticity was different in the two circumstances; either it was opposite ($n = 8$) or plasticity was expressed only during CS-US pairing and not as a change in the FRF ($n = 5$). Such differential expressions of plasticity in the two circumstances were classified as context-dependent effects, whereas changes in training and FRF determination that were in the same direction (i.e., both increases or decreases) were considered to be context independent. Although context-dependent effects occurred more commonly in VE ($7$ of $8, 88\%$) than in AII ($6$ of $11, 55\%$), the differences were not significant ($p > .1$, chi-square).

In general, each cell was idiosyncratic with regard to its details of learning-induced plasticity. Therefore, we present training and FRF data from individual cases in order to illustrate the richness and complexity of the expression of neuronal plasticity. The data from these cells represent the kinds of effects that learning can have on the discharge activity of neurons in auditory cortex.

Context-Independent Plasticity

In order to provide a background for context-dependent effects, we first present an example of context-independent plasticity.

Cell T18A, located in AII, developed a significant decrease in response to the CS during conditioning trials and also exhibited a frequency-specific decrease to this same stimulus in the postconditioning FRF. Because the effects of learning were the same in both situations, the expression of plasticity for this cell was considered to be independent of context.

In this session, unlike all others, the CS was a tonal triad (10, 11, and 12 kHz) instead of a single tone in order to demonstrate that frequency-specific plasticity could develop to complex stimuli. Of greater interest, the FRF revealed that
Figure 6. Frequency receptive field (FRF) stability determination for cells in ventral ectosylvian auditory field (VE). (FRF stability is shown for cells T11D [top] and T13G [bottom], both of which were located in VE. Each data point is the mean value of 15 repetitions. The two FRFs were determined 20 min apart before the start of training. The correlation coefficients for T11D and T13G were .96 and .65 [p < .001], respectively. SP/SEC = spikes per second and PRE-SENS = presensitization.)
Figure 7. Frequency receptive field (FRF) stability data for cells in secondary auditory cortex (AII). FRF stability is shown for cells T8A [top] and T10D [bottom], both of which were located in AII. Each data point for T8A is the mean value of 15 repetitions; for T10D, the mean values were based on 10 repetitions. The correlation coefficient for both T8A and T10D was .88 [p < .001]. SP/SEC = spikes per second and PRE-SENS = presensitization.)
this cell developed a sensitization effect consisting of a large increase in response to all frequencies. Subsequently, conditioning "carved" out of this sensitized response a narrow band decrease that was restricted to the frequencies of the CS triad (10, 11, and 12 kHz).

Neuronal responses to the CS during training are presented in Figure 8. During sensitization trials, there were no significant changes in the evoked response to the CS presented unpaired with the US. During conditioning (CS-US paired), a decrease in response developed beginning in the first five trials. The response to the CS at the end of the sensitization period (32 spikes/s) declined by greater than 50% (to 14 spikes/s) by the end of the conditioning period.

Figure 9 illustrates the FRFs of cell T18A determined throughout the recording session. Prior to training (see Figure 9, Presensitization) it responded at a relatively constant rate of activity (9 spikes/s) across a broad range of frequencies (1–15 kHz). Although there was no obvious change in evoked activity during sensitization training trials (see Figure 8), the postsensitization FRF (see the middle of Figure 9) indicated that the cell had actually developed a general increase in excitability across frequencies (ANOVA, p < .01); that is, it exhibited sensitization. Following conditioning, during which a decrease in evoked activity developed during training trials (see Figure 8), a highly specific and significant (ANOVA, p < .01) decrease in response to the three frequencies that served as the CS was evident (see Figure 9, Postconditioning).

These data revealed that the CS frequency-specific effects of association were in the same direction (decrease) both during CS-US pairing and in the postconditioning FRF, so that the cell's expression of plasticity was independent of context. Also of interest, these data demonstrate that the same neuron may develop both nonassociative (sensitization) and associative (conditioning) effects, so that the classification of cells as either sensitized or conditioned should be made with caution. In the present case, the associative CS specific decrease might not even have been detectable except on a background of increased response to all frequencies that was caused by sensitization.

<table>
<thead>
<tr>
<th>FRF change</th>
<th>Increase</th>
<th>Decrease</th>
<th>No change</th>
<th>Total</th>
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</thead>
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<td>7*</td>
<td>0</td>
<td>11</td>
</tr>
<tr>
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<td>2</td>
<td>0</td>
<td>3</td>
</tr>
<tr>
<td>No change</td>
<td>2</td>
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</tr>
<tr>
<td>Total</td>
<td>7</td>
<td>12</td>
<td>1</td>
<td>20</td>
</tr>
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</table>

* These interactions identify sessions in which context-dependent changes in evoked activity occurred. In each case, the directions of plasticity between the frequency receptive field (FRF) and the conditioned stimulus–unconditioned stimulus (CS–US) pairing are different.

Figure 8. Context-independent plasticity (training data). (This cell [T18A] developed a significant decrease in evoked activity during conditioning. Data are mean values of 10 trials of evoked activity to the first three frequencies used as the conditioned stimulus [CS; 10, 11, and 12 kHz]. Decreases in evoked activity to the CS also developed in the postconditioning FRF [see Figure 9]. SP/SEC = spikes per second, SENS = sensitization, and COND = conditioning.)
Context-Dependent Plasticity

In this section we present data from three cells in which the expression of plasticity was not the same between CS-US pairing and the postconditioning FRF. As noted earlier, such context-dependent plasticity was found in 13 of 19 cells. In 8 cells, the effects were opposite; in 5 cells, plasticity developed to the CS during conditioning, but there was no change at the CS frequency in the postconditioning FRF (see Table 1). We begin with an example of the latter category.

Cell T9D. This neuron, located in VE, had a relatively stable excitatory response to the CS (2.5 kHz) during the sensitization period (see Figure 10A and 10B). Within the first 5 trials of conditioning, this response diminished and then was eliminated completely by the second block of trials (Trials 6–10). The virtual absence of response to the CS was maintained throughout the final 25 trials of the conditioning phase (see Figure 10B, Conditioning Blocks 2–6). During extinction, which immediately followed conditioning, the evoked response returned (see Figure 10B, Extinction Blocks 1–6). Extinction appeared to reverse the effects of conditioning to the level of response observed during the sensitization period.

The FRFs of cell T9D are presented in Figure 11. Before the sensitization period, the cell exhibited weak excitatory responses to several frequencies (e.g., 0.2, 1.2, and 5.0 kHz) and was slightly but not significantly inhibited at the frequency of the CS (2.5 kHz). Following sensitization, the cell was less responsive to the tones, but the differences between the pre- and postsensitization FRFs were not significant (p > .1). In contrast, immediately after conditioning this neuron exhibited significantly increased responses (p < .05) to a broad range of frequencies, with more significant increases (p < .01) evident at frequencies immediately adjacent to that of the CS (i.e., 1.2, 2.0, 3.0, and 3.5 kHz). Remarkably, the cell remained completely unresponsive to the CS.

Following extinction, the increase in evoked activity to non-CS frequencies was greatly reduced. Most important, frequencies adjacent to the CS were affected less than frequencies more distant from the CS (see Figure 11, Postextinction). This maintenance of evoked activity to frequencies close to the CS suggests that even after behavioral extinction, associatively induced frequency-specific changes in the FRF persist.

The lack of evoked activity to 2.5 kHz in all of the FRF tests contrasts sharply with the dynamics of evoked activity to the same stimulus when presented as the CS during training trials. During conditioning, the onset response was first reduced and then eliminated. The evoked response then returned during extinction (see Figure 10). In contrast, there was never any evidence of a response to the CS frequency in the FRF. For this cell, all significant changes in the FRFs occurred to frequencies other than the CS. The physiological plasticity of this cell was categorized as context dependent because the associative effects differed between CS-US pairing and the postconditioning FRF (i.e., there was a decrease in evoked activity to the CS during training and no change in response to the same stimulus in the postconditioning FRF).

Cell T16B. Data from this cell, located in VE, provide another example of context-dependent plasticity. In addition, we had the rare opportunity to record the activity of this cell during a prolonged training session in which further training procedures were initiated. Specifically, discharges of cell T16B were recorded during sensitization, conditioning, and extinction using 7.5 kHz as the CS. At the completion of these tests, the cell’s waveform remained stable, thus allowing for additional training and FRFs. We then conditioned the animal a second time, in the same recording session, using 18 kHz as the CS. The total time required for this procedure was approximately 4.5 hr.

During conditioning with a CS of 7.5 kHz, the cell developed a significant decrease in response relative to sensitization (see Figure 12, Conditioning 1); the decrease was so great that CS-evoked excitation (30 spikes/s over background activity) was changed to inhibition (reflected as −14 spikes/s...
Figure 10. Context-dependent changes in evoked activity (training data). (In the upper panel, peri-stimulus histograms from cell T9D are presented [10 ms/bin]. The onset response that occurred during sensitization [summation of 20 trials] was abolished during conditioning [30 trials], followed by a return to responsiveness with extinction [25 trials]. In the lower panel is an analysis of the dynamics of the onset response [20–70 ms after tone onset] during training. Each data point is the mean value of evoked activity in 5 trials. This analysis revealed that the evoked response evident during sensitization was greatly reduced in the first 5 trials of conditioning and abolished completely in the final 25 trials. Evoked activity then returned within the first 5 trials of extinction. The dynamics of changes in evoked activity are contrasted with a lack of change to the conditioned stimulus in the FRFs [see Figure 11]. SP/SEC = spikes per second, CS = conditioned stimulus, US = unconditioned stimulus, SENS = sensitization, COND = conditioning, and EXT = extinction.)
in the fourth block of conditioning). During subsequent extinction training, the cell once again became responsive to the CS (Extinction 1). This reversal of the conditioning effect was a consistent phenomenon, occurring for all cells so tested ($n = 9$).

During the second conditioning phase (see Figure 12, Conditioning 2), the cell initially was highly responsive to the CS of 18.0 kHz (42 spikes/s in Conditioning 2, Block 1). However, by Blocks 4–6 (Trials 16–30), the rate of activity had declined considerably. In the final 15 trials of conditioning, the response to the CS was no different from background activity. In the subsequent extinction phase (Extinction 2), the conditioning effect was reversed; the cell developed and maintained a significant response to the CS.

These results may give the impression that learning affected the cell in the same manner during training to 7.5 kHz and then 18 kHz (i.e., a decrease in its response to the CS during conditioning followed by a return of the response during extinction). However, as discussed later, the FRF determinations revealed that the dynamics of plasticity in the
two conditioning sessions were different: Conditioning at 7.5 kHz involved a general, nonspecific change in the FRF, whereas conditioning at 18.0 kHz caused a CS-specific change in the FRF.

The general change in the FRF is apparent in the top panel of Figure 13. Prior to sensitization, the cell responded to stimuli from 9 to 22 kHz (Presensitization). Following sensitization, there was a significant increase in evoked activity to stimuli from 15 to 20 kHz. There was no significant change, however, in the response to the CS frequency in the postsensitization FRF (7.5 kHz, labeled CS 1). More to the point, the postconditioning FRF revealed a significant increase in evoked activity to a broad range of frequencies (i.e., learning induced a nonspecific increase in frequency response).

The frequency-specific effect of conditioning can be seen in the bottom panel of Figure 13, which shows the FRFs obtained after extinction at 7.5 kHz and the postconditioning and postextinction FRFs obtained after using 18 kHz as the CS (labeled CS 2). First, the postextinction FRF for 7.5 kHz (Postextinction 1) shows a reversal of the significant increase in evoked activity in the postconditioning FRF for 7.5 kHz (Postconditioning 1 in the upper panel). The high level of evoked activity present in the first postconditioning FRF (Postconditioning 1) was almost completely eliminated in the first postextinction FRF (Postextinction 1). Second, after conditioning at 18 kHz (Postconditioning 2), the increase in evoked activity was highly specific, occurring only to the CS frequency. This extraordinary specificity is in contrast to the nonspecific increase in evoked activity following conditioning at 7.5 kHz. Following extinction at 18 kHz, the associatively induced increase in response to 18 kHz was abolished (Postextinction 2).

This cell developed a decrease in response to the CS both at 7.5 and 18.0 kHz during conditioning. In contrast, responses to the CS frequency in the postconditioning FRF increased in both cases. Hence, the direction of the effects of training was reversed between training and FRF determinations, thereby satisfying the criterion for context-dependent plasticity.

Cell T20C. The final example of context-sensitive plasticity involves an additional manipulation that we used in order to study the persistence of the conditioning effects. In all cells studied with extinction training (n = 9), the changes in evoked activity in the postconditioning FRF were reversed in the postextinction FRF. There are two possible explanations for this effect: (a) The subjects learned that the CS was no longer a predictor of the US (as measured by the extinction of pupillary CRs), and this relearning of contingencies was
context-dependent changes in evoked activity frequency receptive field (FRF) data. (The FRFs of this cell [T16B] were obtained during two consecutive training phases. Data are the mean values of 10 repetitions. In the upper panel are FRFs from the first phase of training [sensitization and conditioning at 7.5 kHz]. In the post-sensitization FRF, there were increases in response only to frequencies from 16 to 20 kHz. In contrast, there were significant increases in evoked activity to numerous frequencies, including the conditioned stimulus [CS] following conditioning [Postconditioning 1]. In the lower panel are FRFs obtained following extinction at 7.5 kHz and then conditioning and extinction using a CS of 18 kHz. The general increase in evoked activity present in the first postconditioning FRF [upper panel] is no longer present in the following extinction [Postextinction 1]. Following conditioning at 18 kHz [Postconditioning 2], there was a significant increase in evoked activity only to the CS frequency. This increase in evoked response was abolished with extinction [Postextinction 2]. The direction of changes in evoked activity in the FRFs—increases in evoked response following conditioning and decreases with extinction—is in the opposite direction of the changes that developed during training [see Figure 12]. SP/SEC = spikes per second, PRE-SENS = presensitization, POST-SENS = postsensitization, POST-COND = postconditioning, and POST-EXT = postextinction.

Figure 13. Context-dependent changes in evoked activity frequency receptive field (FRF) data. (The FRFs of this cell [T16B] were obtained during two consecutive training phases. Data are the mean values of 10 repetitions. In the upper panel are FRFs from the first phase of training [sensitization and conditioning at 7.5 kHz]. In the post-sensitization FRF, there were increases in response only to frequencies from 16 to 20 kHz. In contrast, there were significant increases in evoked activity to numerous frequencies, including the conditioned stimulus [CS] following conditioning [Postconditioning 1]. In the lower panel are FRFs obtained following extinction at 7.5 kHz and then conditioning and extinction using a CS of 18 kHz. The general increase in evoked activity present in the first postconditioning FRF [upper panel] is no longer present in the following extinction [Postextinction 1]. Following conditioning at 18 kHz [Postconditioning 2], there was a significant increase in evoked activity only to the CS frequency. This increase in evoked response was abolished with extinction [Postextinction 2]. The direction of changes in evoked activity in the FRFs—increases in evoked response following conditioning and decreases with extinction—is in the opposite direction of the changes that developed during training [see Figure 12]. SP/SEC = spikes per second, PRE-SENS = presensitization, POST-SENS = postsensitization, POST-COND = postconditioning, and POST-EXT = postextinction.)

reflected as a reversal of the effects of conditioning on the cells; or (2) the effects of conditioning were transient, and the mere passage of time would have also resulted in the diminution of the plasticity established during conditioning. In order to resolve this issue, cell T20C (and three others) were studied after a period of silence equal to the maximum time required for behavioral extinction (20 min). This period of time is referred to as the retention phase. After the end of this phase, a postretention FRF was obtained.

During conditioning, cell T20C, located in AII, developed a small but statistically significant decrease in evoked activity to the CS (see Figure 14). Although it attained significance, it was actually one of the weakest training effects observed among all cells we studied. Nonetheless, this cell serves as a good example of context-dependent expression of plasticity because of the extraordinary contrast between training and FRF effects.

The evoked activity of cell T20C in the FRFs is presented in Figure 15. In the presensitization FRF, the cell responded well to low (< 3 kHz) and high (22 and 24 kHz) frequencies, with some weaker responsiveness to middle frequencies (6–8, 13–15 kHz). Except for a slight increase in responsiveness to low frequencies, there were no changes with sensitization (Post-sensitization). The CS was 14 kHz, which was in the middle range of frequencies used to determine the FRF. From a baseline of weak responsivity to the CS frequency in the pre- and post-sensitization FRFs, the cell exhibited a highly significant increase in evoked response to the CS (14 kHz) in the postconditioning FRF (see Figure 15, Postconditioning). This increase was accompanied by significant increases in evoked activity throughout the entire middle range of frequencies (6–15 kHz), without comparable effects at lower and higher frequencies. The stark contrast between the large increase in the postconditioning FRF (see Figure 15) and the small decrease in response to the same stimulus during conditioning trials (see Figure 14) exemplifies the extraordinary sensitivity of auditory cortical neurons to contextual cues.

Twenty minutes after the postconditioning FRF, responses to the CS frequency (±1 kHz) remained elevated above preconditioning levels (see Figure 15, Postretention). A direct comparison of the FRFs obtained at the beginning (Presensitization) and the end of the recording session (Postretention) is provided in Figure 16. These data illustrate that the two FRF measures are largely overlapping, except for the enhancement of response in the immediate range of the CS frequency. Thus, over the course of the training session, associative learning induced a lasting and CS-specific change in the FRF of this neuron.

Summary of Results

Data were obtained from 20 single units located in the secondary and ventral ectosylvian auditory cortical fields. In each case, a sensitization phase (CS-US unpaired) preceded conditioning (CS-US paired). Thus, each cell served as its own control for nonassociative effects. Significant changes in evoked activity developed in 19 of 20 sessions (8 increases and 11 decreases). The changes in evoked activity developed in an average of 11.7 training trials (7.8 min). Changes in evoked activity usually preceded the development of the pupillary CR (21.9 trials or 14.6 min).

In addition to recording evoked activity during classical conditioning training, we also determined the FRF of each neuron at least four times during the recording session: (a) twice prior to the initiation of training in order to test for the stability of the FRF; (b) immediately following sensiti-
Discussion

The major finding of this study is that learning-induced plasticity of single neurons in auditory cortex can be expressed differently depending on the context in which stimuli are presented. Before elaborating on this point, we present our criteria for distinguishing the effects as both associatively induced and contextually related. Because the major focus of this article is on contextual effects, we do not discuss in detail the finding that learning induces CS-specific changes in the tuning characteristics of auditory cortical neurons; relevant discussion has been presented elsewhere (Diamond & Weinberger, 1986; Weinberger & Diamond, 1987). We do provide a consideration of CS-specific effects in the Appendix.

Following discussion of criteria for associative and contextual effects, we depart from standard practice. The data presented in the Results section include extensive commentary. Therefore, we devote the major portion of the Discussion section to theoretical issues concerning context in the neurobiology of learning and memory. Specifically, we offer a preliminary model that may prove helpful in formulating testable hypotheses regarding the induction and expression of physiological plasticity in learning and memory.

Auditory Cortical Discharge Plasticity Is Associatively Induced

Changes in auditory system-evoked activity during learning may arise from peripheral sources, such as (a) changes in distance between the sound source and ears (Marsh, Warden, & Hicks, 1962); (b) sound shadowing by the pinnae (Wiener, Pfeiffer, & Backus, 1966); (c) contraction of the middle ear muscles (Carmel & Starr, 1963; Galambos & Rupert, 1959); and (d) masking noise produced by the subject’s own movements (Imig & Weinberger, 1970; Irvine & Webster, 1972). In addition, proprioceptive or other feedback from somatic CRs could produce neuronal activity highly correlated with behavioral learning, yielding apparent associative neuronal plasticity.

In this study the problems of stimulus constancy and motor feedback were eliminated by training the animals while they were under neuromuscular blockade. Although somatic CRs...
Figure 15. Context-dependent change in evoked activity (frequency receptive field [FRF] data). (Shown are peristimulus histogram data obtained before and after sensitization, conditioning, and after a 20-min period of silence at the conclusion of training [postretention]. Initially, the cell responded best to low and very high frequencies [e.g., < 3 kHz and 22 kHz and 24 kHz in the presensitization FRF]. In the postconditioning FRF, there was a highly significant increase in evoked response to numerous frequencies from 1 to 15 kHz. In the postretention FRF, the increase in response evident to the middle range frequencies subsided, with the new peak response at the conditioned stimulus frequency [14 kHz, 150 ms/bin].)
Figure 16. Context-dependent changes in evoked activity (frequency receptive field [FRF] data). (In order to visualize the long-term effects of training, the rate of activity in the FRFs of Figure 15 is presented. The presensitization FRF was obtained just prior to sensitization training, and the postretention FRF was obtained at the end of training, 20 min after the postretention FRF was determined. This analysis shows that the FRF of the cell had been changed such that there was a significant increase in evoked activity to the conditioned stimulus frequency at the end of training. The responses to lower frequencies [e.g., 0.1-9 kHz] and many other frequencies were not significantly affected. SP/SEC = spikes per second.)

Discharge Plasticity in Auditory Cortex Is Independent of Arousal

There are two aspects of the general state of excitability of an organism that have been alleged to modify neuronal responses to the CS during conditioning and hence potentially restrict interpretations about learning: tonic arousal and phasic (conditioned) arousal.

Tonic Arousal

Tonic arousal refers to the state of excitability between trials. It has been claimed that following a US, an "arousal ramp" is initiated so that tonic arousal is very high at the time the next trial (i.e., the next presentation of the CS) is given (Nienhuis & Olds, 1978). If true, a change in response to the CS could reflect an increase in tonic arousal. The putative role of a posttrial arousal ramp may be rejected here for several reasons. First, Nienhuis and Olds did not measure arousal level, so the arousal ramp remains speculative. Second, our randomization of intertrial intervals would prevent a putative arousal ramp from producing a systematic increase or decrease in discharge to the CS over trials. Third, previ-
ously we found that auditory cortical discharge plasticity cannot be accounted for on the basis of tonic arousal, as indexed by tonic pupillary diameter during intertrial intervals (Diamond & Weinberger, 1984; Weinberger et al., 1984).

**Conditioned (Phasic) Arousal**

Although sensitization provides for a relative constancy of the general state of behavioral excitability, it does not control for conditioned arousal effects. Conditioned arousal is a large increase in behavioral excitability that occurs when the CS serves as a reliable signal for the presentation of the US. The CS-evoked arousal is an associative phenomenon, because it occurs during the conditioning phase and not during sensitization. However, a critical issue remains unaddressed in conventional learning paradigms: Does discharge plasticity occur as a function of changes in the specific processing of stimulus importance or is the plasticity a by-product of the increase in excitability that occurs during conditioned arousal?

This issue had been addressed by a number of investigators who have suggested that conditioned arousal per se, rather than changes in information processing, may provide the basis for the discharge plasticity that occurs in the auditory system (Hall & Mark, 1967; Kitzes et al., 1978; Miller et al., 1982).

Elsewhere we have explained why these previous studies are inconclusive (Weinberger & Diamond, 1988; Weinberger et al., 1984). We have also demonstrated that any effects of phasic arousal on auditory cortical neurons cannot explain discharge plasticity induced by associative effects (Diamond & Weinberger, 1984; Weinberger et al., 1984). Additionally, phasic arousal cannot account for the fact that learning-induced plasticity in auditory cortex is specific to the frequency of the CS; that is, it is related to the frequency information of the CS rather than to the conditioned arousing properties of the stimulus (Diamond & Weinberger, 1986; Weinberger & Diamond, 1987).

In the present experiment, we have provided novel evidence that auditory cortical discharge plasticity is present even in the complete absence of conditioned arousal. Specifically, the determination of each FRF involved the presentation of a sequence of up to 30 tones, each of a different frequency, the frequency of the CS was embedded in this sequence of tones. During the FRF determination, subjects were largely unresponsive to the entire sequence of tones, as evidenced by a lack of pupillary dilation, which is a sensitive index of arousal (see Figure 4). In no case did the subjects exhibit pupillary dilation to the frequency of the CS in the FRF determination. Yet, even in the absence of a stimulus-evoked change in behavioral excitability, auditory cortical neurons exhibited discharge plasticity. Thus, a behavioral CR and the increase in arousal that may accompany it are not necessary for the expression of auditory cortical discharge plasticity. To our knowledge, our findings provide the first evidence that learning-induced discharge plasticity to the CS is present following training when the behavioral CR is not present.

In summary, we have presented three levels of support for the conclusion that learning-induced plasticity in auditory cortex reflects associatively based changes in neuronal in-

**Context-Dependent Plasticity: Definitions and Criteria**

The issue of context has been investigated extensively in behavioral studies of human and animal learning (e.g., Godden & Baddeley, 1980; Mackintosh, 1985). This inquiry has focused on the extent to which associations other than the CS-US relation are established. For example, pigeons trained in different chambers, distinctive because of different wallpaper patterns, associate an aversive stimulus with the wallpaper pattern, as well as with the specific CS.

In the present experiment, we defined context on the basis of the physical parameters of stimuli presented to the subjects. Two basic contexts are recognized: (a) classical conditioning training and (b) the period during which the FRF was determined (FRF context). The training context was distinguished by the presentation of the CS as the sole acoustic stimulus and the presence of the US. During sensitization the CS and US were unpaired, whereas in conditioning they were explicitly paired on each trial. In contrast, the FRF context involved the presentation of numerous different acoustic stimuli, only one of which was the CS. During the determination of the FRF, the US was not presented.

Because the physical parameters of the CS were identical throughout the recording session, it was possible to directly compare the effects of learning on responses with this stimulus in each context. Differences in response would support an interpretation of contextual influences. The criterion for classifying changes as context dependent was stringent: The cell must have developed a qualitative difference response between the two contexts; quantitative differences would not have satisfied the criterion. For example, a decrease in CS-evoked activity during training and an increase in evoked activity to the CS in the postconditioning FRF would be classified as context-dependent plasticity. In spite of this criterion's stringency, the majority of cells recorded in auditory cortex (68%) satisfied the criterion.

**The Functional Mosaic:**

**Toward a Theory of Context-Dependent Expression of Neurophysiological Plasticity**

The Functional Mosaic concept embodies the view that learning changes the constellation of ways in which a neuron responds to a stimulus in a situation-dependent manner. In
this model, neurons operate within a mosaic of influences; there is no single mode of response on which learning operates or that will be manifest following learning. In Figure 17 we present a simple schema that may help illustrate how contextual variables could affect the expression of learning-induced physiological plasticity. This initial formulation of the model includes the following assumptions.

First, among the multitude of inputs to a neuron, particular inputs may either be expressed (E) or nonexpressed (PE) in different circumstances; expression is defined as the capacity for an input to induce a change in membrane potential at the target neuron.

Second, if associative plasticity (P) develops, it can do so only during CS-US pairing. Plasticity is defined as the capacity for altered response at the target cell. Once the induction of plasticity occurs, it remains in the later determination of the FRF (and for an indeterminate period thereafter). In contrast, if an input does not develop plasticity during CS-US pairing (P), it then remains nonplastic during the FRF determination. The latter assumption rests on the premise that associatively induced plasticity occurs only during the CS-US pairing. Because the sequences of tones used to obtain the FRF were never presented in temporal proximity to the US, they could not have served as a signal for the US.

Given these assumptions, an input may be in one of four possible states: (a) plastic and expressed (PE), (b) plastic but not expressed (P), (c) nonplastic and expressed (PE), and (d) neither plastic nor expressed (PE).

In Figure 17, the state of particular inputs to a neuron during CS-US pairing is indicated by the columns, and the state during FRF determination is indicated by the rows. Intersections denote the state of an input during each of the contexts. For example, in the upper left corner the input is expressed and plastic (PE) in both CS-US pairing and the FRF determination (idented as ‘0’). This combination and all others along the diagonal would not contribute to context-dependent effects because the input state is identical in the two contexts. In contrast, the case immediately below the last example (identified as ‘+’ along the far left column) indicates that the input is plastic and expressed during CS-US pairing (PE), but the plasticity is not expressed during the FRF determination (PE). Because the states of expression in the two contexts are different, this input (and the three others indicated by +) would contribute to context-dependent effects.

The actual mechanisms underlying context-dependent plasticity are currently unknown. Factors that underlie context-dependent plasticity and hence contribute to different functional mosaics need to be identified, characterized, and become part of a testable theoretical framework. At this early stage, we suggest that one factor that might have influenced the expression of plasticity was the state of behavioral arousal. We were able to determine whether the animals were in the same or different behavioral state in the two contexts because pupillary behavior provides a sensitive index of arousal level. Differences in the state of arousal evoked by presentation of the CS frequency in training, as compared with FRF determination, could contribute to a difference in the expression of plasticity.

In Figure 4, representative pupillary records from four different training sessions are presented. The examples illustrate typical pupillary behavior to the CS in the two contexts, that is, during CS-US pairing and during the postconditioning FRF. In each of the examples, presentation of the CS resulted in a large increase in arousal level during CS-US pairing (see the left side of Figure 4), as indexed by the pupillary CR. In contrast, there was no evidence of a comparable pupillary dilation to the CS when it was presented in the FRF context (see the right side of Figure 4). Hence, the behavioral data indicate that the cue value of the CS was different depending on whether it was presented during training (as an arousing signal of US presentation) or during the FRF (as a neutral stimulus).

The context-dependent difference in behavioral excitabil-
ity provides a means to evaluate the nature of information processing in auditory cortex. In spite of the fact that the behavior of the animals indicated that the CS was neutral during the postconditioning FRF, the auditory cortical neurons responded to this stimulus with significantly altered evoked activity. These findings suggest that the changes in CS-evoked activity during learning cannot be attributed solely, if at all, to processes underlying either response initiation or general arousal. Rather, the maintained expression of learning-induced plasticity in the absence of a CR suggests that auditory cortical activity contributes to the storage of information. This interpretation is substantiated by the findings of a retention of the CS-specific FRF changes 20 min after training was concluded (see Figures 15 and 16; see also Figure 3 of Diamond & Weinberger, 1986). Only in the cases in which extinction training occurred did the CS-specific changes in the FRF return control levels (see Figures 11–13; see also Figure 3 of Diamond & Weinberger, 1986, and discriminative plasticity lasting at least 7 days in Oleson et al., 1975).

Within the framework of the Functional Mosaic conception, the data indicate that the constellation of inputs in auditory cortex is capable of rapid reorganization during changes in stimulus contingencies, such as during conditioning and extinction. Moreover, during periods of acoustic stability, such as during the 20-min period of silence (retention test), the circuitry appears capable of maintaining a stability of the recently acquired modifications.

Conclusions

We propose that the induction of plasticity during learning represents the expression of changes in evoked activity specifically for that particular circumstance. In effect, the plasticity that is observed by an experimenter constitutes only a “snapshot” of a multitude of different forms of expression that the induction procedure initiates. This constitutes a neurobiological parallel to the Heisenberg uncertainty principle of quantum physics. That is, the process of obtaining a measurement actually influences the state of the system. In auditory cortex this influence can result in differences in the combination of expressed and nonexpressed inputs, leading to different forms of discharge activity and ultimately to differences in behavioral responses.

A major implication of the Functional Mosaic view is that in order to fully understand the neuronal bases of learning and memory, it is also necessary to determine the extent and nature of contextual influences. Although this conclusion complicates efforts to understand the physiological basis of learning and memory, it may also provide a conceptual means that can encompass the richness and diversity that characterize the foundational processes of perceptual, cognitive, and behavioral adaptation.4

4 Following completion of the writing of this article, evidence for different cellular mechanisms for the induction and expression of physiological plasticity was reported. Muller, Joly, and Lynch (1988) reported that long-term potentiation (LTP), which may be involved in memory storage, in the in vitro hippocampus involves different classes of excitatory amino acid transmitter receptors. The induction of LTP involves N-methyl-D-aspartate (NMDA) receptors, whereas the expression of LTP involves non-NMDA receptors. Of relevance, LTP also develops in the magnocellular medial geniculate nucleus (MGM), which projects to auditory cortex (Gerren & Weinberger, 1983). Therefore, if the distinction between induction and expression of plasticity found in the hippocampus also applies to the MGM, then this nucleus could contribute to auditory cortical plasticity selectively (e.g., during periods when its plasticity is expressed). The extent to which learning-induced plasticity in the auditory cortex may be local or may depend on plasticity established in the MGM is discussed in detail in Weinberger et al. (in press).

References


CONTEXT-DEPENDENT PLASTICITY IN AUDITORY CORTEX


(Appendix follows on next page)
Appendix

Neurons as Adaptive Filters

The major finding we report here is that learning-induced plasticity of single-unit responses in auditory cortex can be expressed differently depending on the context of acoustic stimulation. However, the characteristics of plasticity of the frequency receptive fields (FRFs) were sufficiently intriguing to warrant additional commentary. In this section, we discuss the concept of cortical neurons functioning as adaptive filters. Specifically, we consider the filtering process performed by cortical neurons as a dynamic, experience-based analysis of environmental sounds.

Auditory Cortical Neurons, Adaptive Filters, and Learning

Filters in electronics and communications networks are selective in passing some range of an input parameter (e.g., frequency) while rejecting other values of that parameter. We consider two mutually exclusive classes of filters: nonadaptive and adaptive. Nonadaptive filters are common in electronics, functioning as a constant frequency filter with settings that remain unaltered as the input signal changes. For example, a band-pass filter is commonly used in physiological applications to a restricted range of frequencies. A neuron firing within the filter range has the capacity to exhibit selective changes in their filtering characteristics as a function of some change in input status.

The FRF of a neuron may be viewed as a graphic representation of the filtering characteristics of that cell because, like the input-output plot of an electronic frequency filter, it is a representation of selective output (discharges) as a function of acoustic input (Suga, 1982). When the FRF is systematically altered by learning, it may be regarded as evidence of adaptive filtering.

The general appearance of FRFs of auditory cortical neurons is one of a band-pass filter. That is, there is a restricted frequency range to which the cell is maximally responsive. Frequencies significantly greater or less than the optimal or best frequency are less likely to induce a change in evoked activity. We raise this elementary point about sensory physiology to contrast known tuning characteristics with the novelty of the current findings. Prior to training, the FRFs of the secondary auditory cortex (AII) and ventral ectosylvian auditory field (VE) neurons were unremarkable in that they exhibited the general appearance of normative auditory cortical responses. Narrow frequency tuning in VE (see Figure 6) and broader tuning in AII (see Figure 7) have both been reported previously in sensory physiological studies of the auditory cortex of the cat (Reale & Imig, 1980). The extraordinary finding was that these apparently “normal” frequency responses were subject to extensive and systematic modification during associative learning.

Although the effects of learning on FRFs are discussed in detail in the Results section, we briefly highlight the filtering characteristics of two neurons in order to illustrate the uniqueness of the findings. Cell T18A exhibited an FRF that is typical of cells recorded in AII in anesthetized animals (Reale & Imig, 1980; Schreiner & Cynader, 1984) in that it was broadly tuned, responding to frequencies over a range of 1.5 kHz prior to training (see Figure 9, Presensitization). Three of the frequencies to which it responded (10, 11, and 12 kHz) were used as a complex conditioned stimulus (CS) during conditioning. At the conclusion of the conditioning phase, this cell exhibited a highly unusual if not unique FRF: There was a conspicuous suppression of evoked activity within the central area of maximal excitation (Postconditioning). This suppression was located precisely at the three frequencies that were used as the CS during training. The characteristics of the postconditioning FRF have the appearance of qualities of both band-pass and notch filters; the band-pass characteristic was that there was a lack of evoked activity to low (< 1 kHz) and high (> 15 kHz) frequencies, whereas the notch filter was active at the frequencies of the CS (10, 11, and 12 kHz).

A second example is cell T9A, located in VE (see Figure 11). Prior to training (Presensitization), this neuron was weakly responsive to tones. In spite of the lack of significant evoked activity, we recorded from this unit during training, which allowed for the possibility that learning may induce the expression of inputs that were subthreshold for evoking discharge activity. Following conditioning training (Postconditioning), this did indeed occur; the lack of responsiveness was no longer present as the cell exhibited significant evoked activity to numerous frequencies in the FRF.

The change in responsiveness of cell T9D such that a broad range of frequencies evoked activity suggests that prior to training, there were auditory inputs that were unable to be expressed as discharge activity. These inputs, by mechanisms not yet understood, became expressed as the subject learned that the CS frequency served as a salient signal. Hence, the cell was transformed from exhibiting a complete blockade of frequencies into one that served, at first glance, as a broadly set band-pass filter. However, as with cell T18A, discussed previously, there was a conspicuous lack of evoked activity to the frequency of the CS (2.5 kHz) in the postconditioning FRF.

The notch filter tuning response described in this study has not, to our knowledge, been documented in any previous analysis of auditory cortex. Apparently, notch filter modes, as well as other forms of adaptive filtering, develop during a period of time in which distinct sounds are deemed significant. Because extinction induces a regression of plasticity toward preconditioning levels (see Figure 3 of Diamond & Weinberger, 1986), the selective filtering effects may endure only as long as the sound is a reliable predictor of reinforcement.

Conclusions

One interpretation of these findings is that auditory cortical neurons, at least those in AII and VE, serve as a temporary storage site of the recognition that particular sounds are important. Following extinction, the filter is “reset” to the original pretraining state. The preconditioning or postextinction FRFs of the cells appear to be a basic ground state, possibly determined solely by the density of the local afferentation. Changes in the FRF would then result from a biasing of synaptic strength for those inputs that convey the salient information.

The finding of a basic ground state in which auditory cortical neurons remain constant, in the absence of changes in acoustic salience, has some bearing on techniques for defining the functional characteristics of sensory systems. In a conventional sensory physiology experiment, the animals are usually anesthetized, thereby greatly reducing the chance that newly acquired information will be revealed. Even in sensory physiology studies using awake animals, the absence of reinforcement and the presentation of a multitude of neutral stimuli virtually eliminates the possibility of associations occurring between particular acoustic stimuli and reinforcement. It is therefore most likely that this type of experiment characterizes the static components of what is otherwise a highly dynamic system. In order to gain a complete understanding of the involvement of auditory cortical fields in learning and memory, the biophysical mechanisms and behavioral variables underlying the static and dynamic properties of these cells need to be determined.