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Modification of Unit Discharges in the Medial Geniculate Nucleus by Click–Shock Pairing

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The present experiment was concerned with some discharge properties of single neurons in the medial geniculate body of the locally anesthetized paralyzed cat. The effect of pairing clicks with paw shock upon discharge rate and pattern was of particular concern. Twelve neurons obtained from 11 cats were studied exhaustively for periods up to 4 hr. Under control conditions, rate stationarity for both spontaneous and click-evoked activity was found in only 3/12 of the units. Click–shock pairing produced rate changes in 10/12 of the cells; an increase in rate predominated. The pattern of discharges was altered in 9/12 of the cells as a consequence of click–shock pairing. Specifically, the initial short-latency discharge was modified; there was a reduction in the proportion of spikes in the first peak to the total number of spikes in the poststimulus time histogram. This reduction was not merely a consequence of increases in over-all rate of discharge. In the case of one cell which was inhibited rather than excited by click stimulation, the click–shock pairing resulted in a reduction in the duration of inhibition. Control findings indicated that the pattern modifications were not due to a change in stimulus intensity, the unconditioned effects of the shock itself, or to an increase in arousal level.

Introduction

The role of sensory systems in behavioral plasticity has not yet been delineated. Whether they serve only to process stimulus information in an invariant, albeit elegant, manner or additionally exhibit dynamic properties critical to behavioral change remains an open question. In regard to the latter issue, negative findings have been reported for the somatosensory system during habituation of the flexion reflex. (11) and for the auditory system during habituation of the orienting reflex (13). Less is known for other behavioral situations, but if sensory processing is related in a dynamic fashion to changing behavior, it would follow that this would be re-

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vealed in the discharge properties of individual sensory system neurons. In support of the dynamic role, it has been found that the pairing of flashes with somatic shock systematically alters the flash-evoked discharge patterning of cells in the visual cortex (8). Similarly, the pairing of different types of visual stimuli results in a modification of unit response to the first of the pair in the lateral geniculate body (5). Such conditioning effects suggest at the very least that the response of visual system cells in the forebrain may be controlled not only by stimulus parameters but also by some type of associative factor. The purpose of the present study was to ascertain whether neurons in the thalamic component of the auditory system (medical geniculate nucleus) are also subject to such modification. Three discharge characteristics were investigated: stationarity; rate; and pattern as evidenced by the poststimulus time histogram (PSTH).

Methods

Subjects and Surgical Procedures. The subjects were 11 adult cats weighing 2.5–3.5 kg. To be included in the study they were required to give an orienting response to a sudden sound. All surgical and experimental procedures were carried out with the cat enclosed in an acoustically dampened room (IAC No. 1202). The animals were initially anesthetized with an intravenous injection of a short-lasting barbiturate, sodium thiamylal (Surital). The left femoral vein was incannulated to allow for subsequent doses of Surital required to maintain surgical anesthesia and for the later administration of gallamine triethiodide (Flaxedil). Atropine was administered (1 mg, ip) to prevent respiratory congestion. An endotracheal tube coated with a long-lasting local anesthetic (Zyjectin) was inserted under laryngoscopic control. The animal was mounted in a stereotaxic instrument with hollow ear bars through which acoustic stimuli were presented. The cranium was exposed and cleared, a trephine hole placed to allow access to the medial geniculate body, and the dura mater cut and retracted. Stainless-steel screws were placed in burr holes over the suprasylvian gyrus and frontal sinus to provide a monitor of the EEG. Shock electrodes were attached to the hind leg contralateral to the MGB trephine hole. All wound edges and pressure points were heavily infiltrated with Zyljectin and general anesthesia was discontinued. When the animal exhibited spontaneous movements, the endotracheal tube was connected to a respirator and 2 ml of Flaxedil injected. Paralysis was maintained throughout the experiment with gravity infusion of Flaxedil diluted to 10% with dextrose saline solution. Body temperature was maintained at 37 C by a circulating water pad and monitored with a rectal thermometer.

Stimulation and Recording. Acoustic stimuli consisted of clicks produced by 0.5-msec square waves from a Tektronix 162 pulse generator and fed

through Hewlett–Packard 350D attenuators to Grason–Stadler D30 earphones mounted on the ear bars. Click intensity was approximately 85 db re: .0002 μbar. Paw shock consisted of a 50-msec train of 5-msec pulses at the rate of 100/sec produced by a Grass S-8 stimulator. Shock intensity was set to produce a slight flexion response prior to paralysis. Medial geniculate (MGB) units were recorded with glass-coated tungsten microelectrodes of 3–10 megohms impedance and were amplified by conventional means. Further details are provided in a previous report (4). Unit spikes were passed through a Schmitt trigger which provided standard pulses to a LAB-8 computer. Exceptional care was given to monitoring unit waveform and amplitude. Only neurons whose waveform was constant and whose amplitude was greater than three times that of the background activity were studied.

Experimental Procedure. Units were located by presenting clicks occasionally as the microdrive was lowered. After a cell was isolated, the following paradigm was instituted. Cell discharges were recorded during three conditions: click stimulation of the right ear; left ear; and in the absence of stimulation ("spontaneous"). Within each condition, stimulation was presented in blocks of 48–100 stimuli. For spontaneous activity, synch pulses were recorded for purposes of data analysis but clicks not presented. The three conditions were presented alternately (e.g., right, left, spontaneous), the entire sequence being repeated several times until sufficient preshock data has been gathered. A few cells also received binaural stimulation. After the recording of the preshock control data, paw shock was paired with click stimulation of either the right or left ear chosen at random. Clicks were presented every 1.3 or 1.6 sec; the click–shock interval was 300 or 600 msec, respectively. These parameters were constant for a given unit. The sequence of click and spontaneous blocks was continued as before until sufficient data had been gathered, or more usually until the unit had been lost. For some units shock was omitted from every fifth click–shock block to provide an estimate of the effects of click–shock pairing in the absence of shock itself. In a few cases, units were held long enough to permit discontinuation of shock pairing, and return to the nonshock control paradigm.

In summary, unit discharges were recorded during right, and left click presentation and the absence of stimulation. This sequence was repeated during three experimental periods: preshock; during the pairing of shock with either right of left ear clicks; and after the discontinuation of shock. This procedure yielded only 12 acceptable cells from 11 cats. These were recorded for periods of 2–4 hr each. Many other cells were lost after an hour or so, before sufficient shock-pairing data had been acquired for statistical analysis and accordingly none of their data are reported. While the

experimental procedure was quite protracted and limited the number of cells in this study, we believe that the issue of discharge modulation required this paradigm, and further that the intensive study of these 12 cells has yielded a finding of general interest.

Histology. At the termination of an experiment, a marking lesion was made by passing 3–5 mcoul of current through the microelectrode and the animal was killed with an overdose of Surital. The animal was perfused with saline solution followed by 10% formalin injected through the heart. Serial coronal sections of 50μ thickness were prepared on a freezing microtome and alternate sections stained with cresyl violet. Sections were photographed at 10× magnification and cell locations established.

Data Analysis. Synch pulses and unit discharges were analyzed by a LAB-8 computer which transferred these data to digital tape preserving all interspike intervals within an accuracy of 100 μsec. This information was subjected to further analysis off-line, as will be described. An earlier report (4) contains a discussion of issues related to our statistical evaluation of unitary data.

Results

Locus of Cells. All of the units reported here were within the parvocellular portion of the MGB. However, they cannot be ascribed with certainty to the dorsal or ventral subdivisions as described by Morest (7) for reasons described previously (4).

Stationarity. By "stationarity" we refer to the absence of significant changes in rate over time for both stimulated and spontaneous conditions. It was computed separately for the preshock (PS) and during-shock (DS) periods. The number of spikes between synch pulses was computed for each data block (i.e., 48–100 consecutive stimuli or synch pulses). The sequential distribution of these spike counts provides a display termed the "serial spike-counts histogram" (SSH) that depicts discharge rate over time (Figs. 1, 2, and 3). Each SSH is comprised of all data blocks within a given condition of stimulation (e.g., right ear) even though that condition alternated with others (e.g., left ear, spontaneous) during actual recording. A one-way analysis of variance was computed for each SSH. If this analysis was significant at the 0.05 level or less, the unit was considered nonstationary for the condition tested; if not, it was considered stationary.

During the PS period 7 of 11 cells (64%) were stationary for the spontaneous condition,[2] and 4 of 12 (33%) during click stimulation. Only three cells were stationary for both the spontaneous and click-stimulated

[2] Spontaneous data for one cell was unavailable.

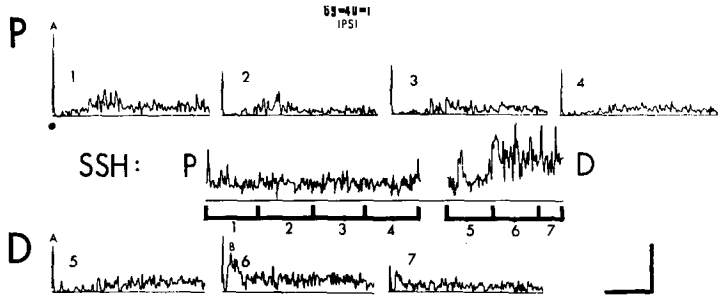

FIG. 1. Rate and patterning of discharges for a cell receiving ipsilateral click stimulation for the preshock period (P) and during the period this stimulation was paired with shock (D). In this and subsequent figures the dot indicates the time of click presentation. In this and Fig. 2 the rate is displayed as the serial spike-counts histogram (SSH) (see text) and patterning is displayed as poststimulus time histograms (PSTH). SSH and PSTH from corresponding data blocks are identified by the same numerals. This unit yielded the least amount of data in the entire sample, and was lost during block 7 as seen in the truncated SSH and half-amplitude PSTH. There was a significant increase in rate which developed during the D period. The pattern was also modified, the proportion of spikes in the initial peak (A) being reduced; this was a consequence of the increased rate because the absolute number of spikes in the peak was not reduced (compare PSTH 5 and 6). The increase in rate was allocated almost entirely to the B peak and background following it. There were 50 click presentations per data block. Calibrations: SSH, 26 spikes; PSTH, 29 spikes and 398 msec.

conditions. Thus, in the absence of designated disturbance of the animal, a significant proportion of MGB cells are not stationary. During the DS period only four units were stationary for the spontaneous condition; the click-stimulation percentage was not changed.

Effects of Shock-Pairing Upon Rate. The effects of shock pairing upon discharge rate were analyzed by first calculating the mean rate per data block and then comparing these rates for PS and DS data blocks by the Mann–Whitney *U* test. In order to eliminate the effects of shock artifact, the rate calculations were limited to the interval prior to shock onset (i.e., either 300 or 600 msec).

Shock pairing significantly altered the discharge rate of 10 cells (83%); six showed increases for all conditions (i.e., right, left, and spontaneous), one showed a decrease, and three had mixed results. If one sums over all stimulus conditions for the 12 cells, there were 34 tests for rate effects.[3] Of these, 24 (71%) were changed, 18 (75%) showed an increase, and six a

[3] There should have been 36 tests (three conditions × 12 units), plus two for two cells given binaural stimulation, making 38. However, insufficient spontaneous activity was recorded for two cells, and two others were activated only by stimulation of one ear.

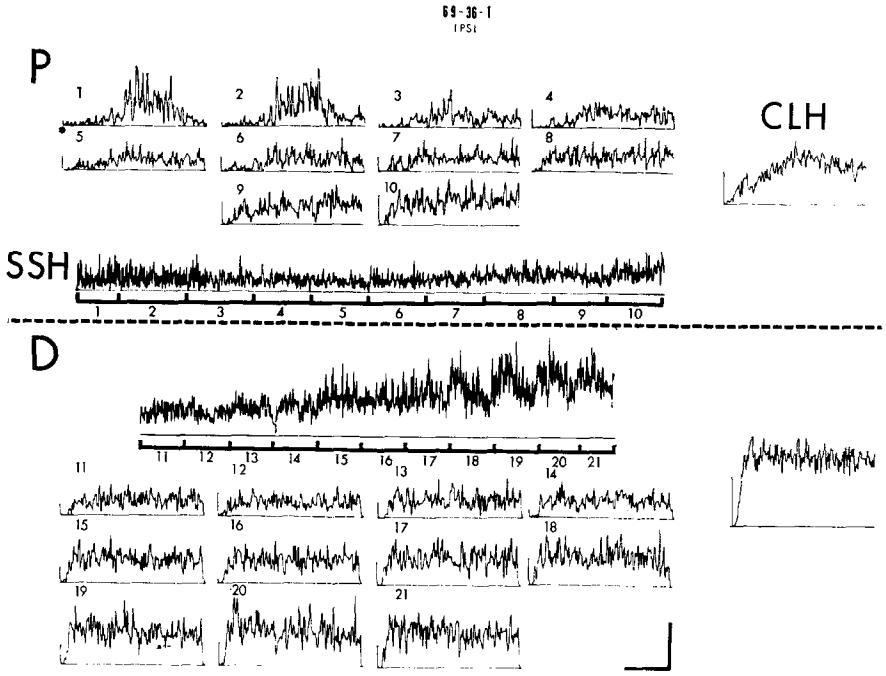

FIG. 2. Rate and patterning of a cell receiving ipsilateral click stimulation which was not paired with shock; shock was paired with contralateral stimulation (not shown). This cell was inhibited by clicks, and the duration of this inhibition was attenuated during the D period (compare CLH for P and D). There was also a pronounced increase in rate (SSH, D) which developed over time. Each data block consists of 100 click presentations. Calibrations: SSH, 26 spikes; PSTH, 17 spikes; CLH, 45 spikes; 210 msec.

decrease in rate. Thus, whether calculating on the basis of cell types or number of statistical tests across cells, it is clear that click–shock pairing is highly likely to change the rate of discharge and that the change is predominantly an increase in rate (Figs. 1, 2).

Pattern Analysis. An impression of the effects of shock pairing upon the pattern of unit discharges may be gleaned by inspection of the cumulative poststimulus time histograms (CLH), i.e., the sum of all block PSTH within a PS or DS condition (Fig. 2). However, this approach may be misleading because the CLH do not reflect variability within each condition. An inspection of the individual block PSTH does provide this information (Fig. 1) but the mass of data may be too large to permit easy visual comprehension (Fig. 2). This problem is intensified by the fact that the rate may not be stationary. For example, inspection of the height of a peak in the PSTH may not be an adequate index. While its number of

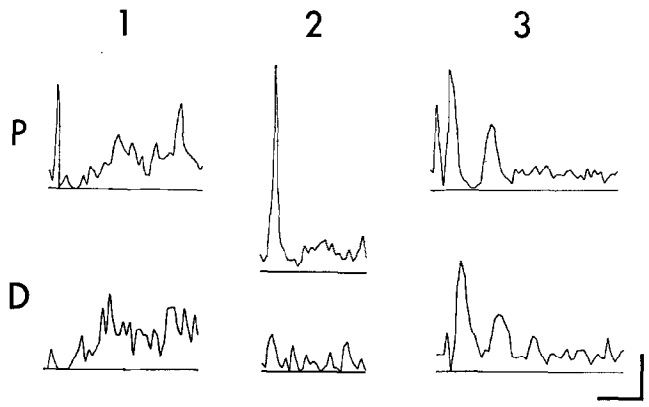

FIG. 3. The effects of shock pairing on patterning of discharges for three units (1, 2, 3) receiving contralateral stimulation. In each case this stimulation was paired with shock (D). Cumulative PSTH (CLH) for all blocks in the P and D periods are presented. Note the reduction in the initial peak relative to the rest of the histogram. Cells 1 and 2 also suffered a reduction in the absolute number of spikes in the initial peak. Note that in this and Fig. 4 the histograms in the P and D periods have different scales to permit plotting the background levels at approximately equal amplitudes. The cumulative PSTH are comprised of the following numbers of click presentations: No. 1, P = 232, D = 120; No. 2, P = 563, D = 563; No. 3, P = 300, D = 135. Calibrations: No. 1, P = 13, D = 7 spikes, and 96 msec. No. 2, P = 25, D = 13 spikes, and 121 msec. No. 3, P = 52, D = 27 spikes, and 130 msec.

spikes may remain constant, other portions of the PSTH may change drastically. Thus, the peak in question actually would have been altered with respect to the total discharge after click stimulation. Therefore, we sought an objective index of the effects of shock pairing upon the pattern of discharges, one which would also be unaffected by rate nonstationarities.

One solution to this problem is to obtain a descriptor of relative pattern by calculating the ratio of spikes in a given peak to the total number of spikes in its PSTH. This solves the problem of having the absolute peak apparently change merely as a function of an over-all change in rate even though the same relative pattern is maintained. On the other hand, the relative measure would indicate peak changes if there were either no change in absolute spikes in the peak while the over-all rate increased or an actual reduction in spike counts in the peak without regard to over-all rate. Analysis of the absolute number of spikes in the peak resolves this second problem and additionally provides interesting information in its own right. Therefore, both types of pattern analysis were used in this study.

In order to determine statistically whether the pattern had changed from the PS to the DS period, peaks were selected by inspection of the CLH. For the relative analysis, the number of spikes in the peak bins was pro-

vided by the computer, and the ratio of spikes in the peak to total spikes in each block PSTH was computed. The ratios for blocks within the PS and DS conditions were compared by a two-tailed Mann–Whitney test (10) to determine the effects of shock pairing. For the absolute analysis an identical analysis was performed on the absolute number of spikes in a selected peak.

The characteristic pattern, as revealed by the CLH, consisted of a short-latency peak or inhibition (10–75 msec.) which might be followed by one or more peaks of up to 300-msec latency. Although the pattern analyses were performed upon all peaks, only the initial short-latency peak (or trough) was found in all units. The following analysis is confined mainly to the effects on these short-latency components of the PSTH.

Nine of the total sample of 12 cells (75%) exhibited significant pattern changes ($P = 0.05$ or less). In seven cells the ratio of spikes in the initial peak was attenuated (Figs. 3, 4).

The eighth cell was inhibited by the click and this inhibition was also attenuated (Fig. 2). The ninth cell showed an enhancement of the first small peak which had a 20-msec latency; its second peak at 55 msec was unaffected but the third large peak at 100 msec was significantly attenuated. For the three cells not exhibiting a relative pattern change, two were recorded from the same cat. The third exhibited reduction in the initial peak after, not during, shock pairing ($P < 0.10$), not significant by our criterion).

In order to determine if reductions in the initial peak (trough) were merely a function of increases in rate consequent to the introduction of shock, the absolute spike counts for the PS and DS period were subjected to Mann–Whitney tests. Five of the eight cells exhibiting a relative attenuation of the initial peak (trough) also had an absolute decrease in initial peak spikes (Figs. 3, 4A "Sh"). Therefore, the effects of shock upon pattern cannot be attributed simply to an increase in over-all rate during the DS period. The rate increase was probably critical for three cells which did not suffer an absolute reduction (Fig. 1).

An obvious interpretation of these findings is that the pattern changes were due simply to the unconditioned effects of the shock itself. Two types of observation suggest that this is not the case. First, if the pattern changes were due to the direct effect of shock, then they ought to be in evidence immediately, during the first DS block. However, several cells revealed a systematic trend of pattern change over time (Fig. 5). Second, the pattern changes occurred in the absence of shock, under the following conditions. (a) Three cells received "test" blocks in the DS period, consisting of presentation of stimulation to the ear ordinarily paired with shock, but with shock omitted. In all three cases, the initial peak attenua-

tion was present in this test condition for the DS period. (b) One cell exhibited the peak attenuation for stimulation to the nonshocked ear; there was no effect for the shocked ear. (c) Five cells exhibited the initial peak attenuation for stimulation to the nonshocked ear as well as for the shocked ear (Figs. 2, 4A). (d) One of these five cells also was recorded during the postshock period; the peak attenuation for the nonshocked ear was still present.

It might also be argued that the peak attenuation was due to a reduction in stimulus intensity. The animals were well paralyzed, eliminating usual sources of stimulus variability, but a build-up of pressure in the bulla which has the effect of attenuating stimulus intensity has been noted (A. Starr, personal communication). Such a pressure change might have

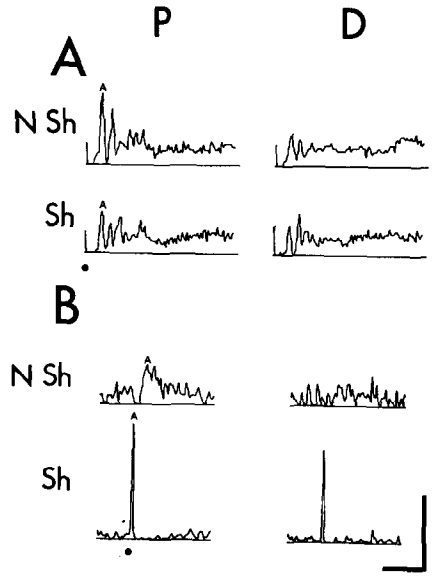

FIG. 4. Pattern of discharges for two cells (A and B) during the P and D periods for both the shocked (Sh) and nonshocked (N Sh) ears. A: This cell exhibited a significant reduction in the proportion of spikes in the initial peak (A) in the D period for both the shocked and nonshocked ears. There was also a significant reduction in the absolute number of spikes in the peak for the shocked condition. The brief initial inhibition was unaffected. B: A unit which exhibited a reduction in both the absolute and relative number of spikes in the initial peak (A) during the nonshocked condition. There was no significant effect on the initial peak for the shocked condition. A is comprised of the following numbers of click presentations: N Sh: P = 480, D = 760; Sh: P = 440, D = 875. B is comprised of these numbers of click presentations: N Sh: P = 300, D = 160; Sh: P = 250, D = 100. Calibrations: A, N Sh: P = 120, D = 190; Sh: P = 110, D = 208. B, N Sh: P = 125, D = 50; Sh: P = 50, D = 17. Time base = 200 msec for both A and B.

coincided with the DS periods in this study. However, the following observations indicate that this effect was not the cause of the initial peak attenuation. Three cells had a peak effect for stimulation to only one ear during the DS period: for two cells the pattern effect occurred only for the ear paired with shock, not for the nonshocked ear; in one cell, the effect was present only for the nonshocked ear, not the shocked side (Fig. 4B). One cell which had the peak attenuated for the DS period lost this change in the postshock period, incompatible with a gradual pressure build-up from the time of paralysis.

Discussion

The present findings demonstrate that nonstationarity is characteristic of many MGB cells studied in quiescent (preshock) conditions. The introduction of shock generally causes an increase in the rate of firing for both spontaneous and click-stimulation conditions. Both of these findings are in agreement with previous observations in which paw shock was introduced

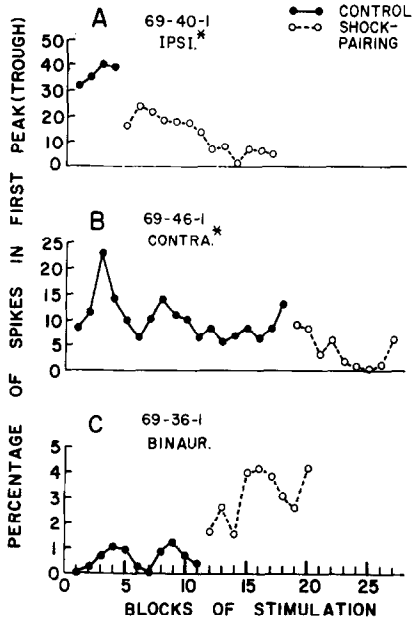

FIG. 5. The development of pattern changes over time for three units. Asterisks denote that results plotted were for the ear paired with shock. The percentage of spikes in the initial peak was reduced during shock pairing for the unit data plotted in A and B. The unit for the C data was inhibited by the click, and the proportion of spikes in the initial trough was significantly increased. Note the absence of a sudden change and the gradual development during shock pairing.

but not paired with acoustic stimulation (4). The rate increases in particular would seem to be a function of some general variable such as an increase in arousal level as they occur in conditions of both paired and unpaired shock. A previous report of the effects of giving shocks during the presentation of 1- 2-min noise stimulation found no effects upon either the background or evoked multiple unit level in the medial geniculate body (2); however, a profound attenuation was reported for the inferior colliculus. Differences in experimental paradigms, stimulation, and recording techniques preclude direct comparisons with the present study so that the conflicting results may be more apparent than real.

The present observations offer no pertinent information for the mechanism underlying the rate increases except to strongly suggest they were not due to changes in the stimulus. Clearly, there is no reason to believe that increases in discharge rate are limited to the MGB or that they are intrinsic to it rather than imposed from other neural systems, or reflective of possible increases in ascending auditory activity (9). Determination of the most peripheral level at which they occur will clarify this issue and also settle the problem of stimulus constancy conclusively.

The pairing of clicks with shocks resulted in an attenuation of the initial short latency discharge. This effect was not a function of possible direct effects of the shock upon MGB cells for it was also found when shock was not actually present, and could develop over time. Neither could the attenuation be attributed to a reduction in stimulus intensity. Therefore, it appears that the attenuation of the initial discharge is due to some other aspect of the introduction of shock. One possibility is a general arousal effect, but this also seems unlikely because a previous study of arousal effects of nonpaired shock upon MGB discharge patterns revealed that the initial short-latency discharge was unaffected (4). Thus, it appears that the pattern change was due to the contingency between click and shock presentation. The formal similarity of the pairing paradigm used here to Pavlovian differential conditioning raises the question of whether conditioning changes of MGB discharge patterns were correlated with stimulus control of behavior. However, the present study did not record or measure available (i.e., autonomic) behavior and, therefore, relationships to behavioral conditioning and discrimination must be purely conjectural and remain so until the appropriate experiments are performed.

The pattern changes do have significance in their own right. Attenuation of the relative initial discharge was due apparently to the reduction in absolute peak spike counts in some cells, and to an increase in overall rate in others. While two different mechanisms might account for these findings, it is clear, nevertheless, that the initial discharge became "submerged" to some extent in the background firing of the PSTH. This implies a modifi-

cation of transmission to the auditory cortex, assuming that the time-locked firing of MGB cells which is revealed to the experimenter in the PSTH is also critical to the auditory cortex.

It is generally held that the initial short-latency evoked activity in sensory systems is relatively impervious to changes in organismic variables, particularly in contrast to longer-latency events which are highly labile. For example, the initial wave of the click-evoked potential in the MGB or auditory cortex is altered minimally or not at all while later waves change drastically during desynchronized sleep (1), and between wakefulness and slow-wave sleep (12) even while lower levels of the auditory system exhibit constancy (14). Parallel effects of arousal upon MGB unit discharge patterns has been reported previously (4). On the other hand, considerable variation in both the early and late waves of the click-evoked potential during fear conditioning have been reported for the MGB and auditory cortex but only the later waves were systematically related to behavior (3, 6). In the present case, shock pairing also altered the short-latency evoked activity, although the effect seemed more systematic than in the evoked potential studies (3, 6). While direct comparisons of field potential and single unit studies should be made with caution, these findings do suggest that short-latency acoustically evoked activity in the auditory forebrain is affected to a greater degree under conditions of click–shock pairing than for general changes in arousal level.

The striking attenuation of the initial discharge was complemented by the reduction in inhibition in a cell not excited by the click. Although more data are needed, it seems reasonable to hypothesize that shock pairing affects the time domain immediately after stimulus presentation without regard to the specific nature of the cell's response. In any event, it is clear that fairly drastic changes in MGB unit discharge pattern can be produced by click–shock pairing. Thus, auditory system cellls, like some neurons in the visual forebrain (5, 8), exhibit dynamic properties which qualify them as candidates for some critical role in the neural bases of behavioral change. However, correlative and ultimately causitive relationships remain to be demonstrated.

References

1. Berlucchi, G., J. B. Munson, and G. Rizzolatti. 1967 Changes in click-evoked responses in the auditory system and the cerebellum of free-moving cats during sleep and waking. *Arch. Ital. Biol.* **105**: 118–135.

2. Galin, D. 1965. Background and evoked activity in the auditory pathway: effects of noise-shock pairing. *Science* **149**: 761–763.

3. Hali, R. D., and R. G. Mark. 1967. Fear and the modification of acoustically evoked potentials during conditioning. *J. Neurophysiol.* **30**: 893–910.

4. IMIG, T., N. M. WEINBERGER, and I. S. WESTENBERG. 1972. Relationships among unit discharge rate, pattern, and phasic arousal in the medial geniculate nucleus of the waking cat. *Exp. Neurol.* **35**: 337–357.
5. LINDSLEY, D. F., K. L. CHOW, and M. GOLLENDER. 1967. Dichoptic interactions of lateral geniculate neurons of cats to contralateral and ipsilateral eye stimulation. *J. Neurophysiol.* **30**: 628–644.
6. MARK, R. C., and R. D. HALL. 1967. Acoustically evoked potentials in the rat during conditioning. *J. Neurophysiol.* **30**: 875–892.
7. MOREST, D. K. 1964. The neuronal architecture of the medial geniculate body of the cat. *J. Anat.* **99**: 611–630.
8. MORRELL, F. 1967. Electrical signs of sensory coding, pp. 452–468. *In* "The Neurosciences." G. C. Quarton *et al.* [Eds.]. Rockefeller Univ. Press, New York.
9. PODVOLL, E. M., and S. J. GOODMAN. 1967. Averaged neural electrical activity and arousal. *Science* **155**: 223–225.
10. SIEGEL, S. 1956. "Nonparametric Statistics for the Behavioral Sciences." McGraw–Hill, New York.
11. SPENCER, W. A., R. F. THOMPSON, and D. NEILSON, JR. 1966. Alterations in responsiveness of ascending and reflex pathways activated by iterated cutaneous afferent volleys. *J. Neurophysiol.* **29**: 240–252.
12. TEAS, D. C., and N. Y. KIANG. 1964. Evoked responses from the auditory cortex. *Exp. Neurol.* **10**: 91–119.
13. WEINBERGER, N. M., D. A. GOODMAN, and L. M. KITZES. 1969. Is behavioral habituation a function of peripheral auditory system blockade? *Commun. Behav. Biol.* **3**: 111–116.
14. WICKELGREN, W. O. 1968. Effect of state of arousal on click-evoked responses in cats. *J. Neurophysiol.* **31**: 757–769.