Response of Single Medial Geniculate Units to Repetitive Click Stimuli

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ABSTRACT

VERNIER, VERNON G. AND ROBERT GALAMBOS. (Walter Reed Army Med. Ctr., Washington, D. C.) Response of single medial geniculate units to repetitive click stimuli. Am. J. Physiol. 188(2): 233-237. 1957.—The capacity of single units isolated in the medial geniculate of unanesthetized, paralyzed cats to respond to clicks delivered to the ear at rates between 1 and 200/sec. is described. Most units respond one-to-one at low click rates. When the interval between stimuli is shortened some stimuli fail to evoke response, and when it is made very brief the unit may respond only to the first click in the train. The click frequency at which a unit responds to 50% of the stimuli is reasonably stable, but can be modified by the administration of morphine.

The two ideas behind the studies reported here were first to add to the available (1, 2) information upon the behavior of single medial geniculate units, and second, to discover what if any changes in that behavior occur when active pharmacological agents are introduced into the bloodstream of the animal. To these ends the capacity of the units to respond to clicks presented at low, intermediate and high rates was determined. As will be developed, the units fail to follow successive stimuli at certain critical rapid rates in the normal animal, and this point of failure may be significantly altered by pharmacological agents. It is well known that a need for quantitative methods for the assay of the effects of drugs upon the nervous system exists, and our success in using single units for this purpose, while limited, may be of interest.

METHODS

Forty-six young adult cats were used in this study. After preparation under ether, they were continued on gallamine (Flaxedil) and local procaine. Unilateral pneumothorax was performed to decrease brain pulsation attributable to respiration. Rectal temperature was maintained in the vicinity of 38°C.

The cat was located in an electrically shielded, sound-deadened room into which respirator tubing, micromanipulator hydraulic coupling and polyethylene intravenous cannula for injections passed from the outside. For isolating units machine-drawn glass micropipets, measuring 0.5–1.0 microns at the tip and filled with 3 M KCl according to the method of Tasaki (3), were used. Their resistance as measured with a high impedance vacuum tube voltmeter in KCl usually ranged from 5 to 20 megohms, averaging about 10.

Auditory stimuli consisted of trains of clicks at various rates, white noise and 1 kHz tones. The clicks were generated by delivering 0.1 msec. square waves at rates of 0.1–200/sec. to a power amplifier into PDR-10 earphones. The sounds could be attenuated in 1 db steps below the maximum; with this equipment the normal human threshold for the click is at approximately –85 db. The sound path to the cat was completed by a 1-cm length of polyethylene tubing coupled to hollow stereotaxic ear bars; these were inserted into the external canal and held the head in a stereotaxic instrument. The ear bars did not penetrate the ear-drum. Calibrations of the click made with a microphone in place of the cat’s head showed the major acoustic perturbation to last less than 0.5 msec for –50 db clicks. Ordinarily, clicks weaker than –50 db were used. The micropipet support permitted millimeter movements in both the antero-posterior and the medio-lateral directions, and was advanced by a hydraulic system operated from outside the experimental room. The electrode could be advanced in 5 micron steps in the Horsley-Clarke coordinates.

A 15-cm silver wire led from the pipet to a cathode follower, which coupled to a Tektronix amplifier (model 122). Recording from each round window was made with similar amplifiers. A common reference point for all electrodes was located in the neck. A four-beam oscilloscope was used for recording the traces, being photographed usually at 250 mm/sec. on moving 35-mm film.

During each experiment the response from at least one round window was monitored throughout to estimate stimulus effectiveness and physiological condition of the animal. The microelectrode was always slowly advanced in the vertical plane toward the...
medial geniculate, while clicks at 5/sec., 10–20 db above the thresholds for round window N1 responses were being presented. When an auditory unit was encountered, its response to clicks, tones and noise, the ear best responded to, the latency to clicks, etc., were then approximated. Threshold for firing to clicks at 5 or 10/sec. then was determined, the intensity was set at 10–20 db above this value, and the responses to at least 50 clicks at repetition rates between 5 and 200/sec. were photographed. The first of several doses of morphine sulfate (1.0 mg/kg as a 4.0-mg/cc solution in 0.9% NaCl and followed by 1 cc of saline to flush the catheter) then was administered slowly over a 60-second period, and the responses to repetitive clicks were photographed once more. This procedure was repeated until the unit was lost. The coordinates of the electrode position then were read, and the cat was killed. On some occasions the brain was perfused and saved for histological control.

**RESULTS**

**Recording of Auditory Units.** The majority of the recordings of single auditory units discussed in this paper were obtained from anterior levels of the medial geniculate body, chiefly in the A7 frontal plane of the stereotaxic coordinates. Most effective for vertical approach was A7, L8, although recordings at A6 and A5 were on occasion successful. As reported earlier (4), it is in the pars principalis (parvicellular portion) of the nucleus, containing cells 20–40 microns in diameter, that units responding to sounds are most likely to be found.

The three types of spike potentials seen in this study and recorded in figure 1 correspond with what previously has been described (5, 6). The first type, small negative spikes (fig. 1A), frequently was seen when there was evidence that the electrode tip had broken and was somewhat larger than its initial 0.5–1.0 microns. On occasion several such spikes of different heights occurred in the same sweep and a large micromanipulator adjustment proved necessary to alter the picture. Our observations agree with those previously published, in associating these small negative spikes with probable extracellular recording from cells or fibers.

The second type of spike, initially positive and biphasic (fig. 1B), was the one most frequently seen. These are probably recorded either directly from the cell surface or with minimal penetration.

The mainly positive larger action potentials of the third type (fig. 1C), which were of the highest voltage, are probably derived from the inside of cells or fibers.

The unit pictured in figure 1 changed progressively from type 1 response to type 3 during the course of the experiment. Typically, as in this instance, a unit would record first as a small negative spike; later, presumably as the electrode advanced, an early positive notch appeared which grew in size until it exceeded the subsequent negative component by several times. Thereafter, one of several changes might occur. Most often, the biphasic positive potential changed slowly into a monophasic positive potential, as in figure 1. This latter might slowly grow even further in amplitude, but if this occurred suddenly the unit displayed an injury discharge and disappeared. On the other hand, the potential might return to the biphasic form. If the unit suddenly disappeared without the injury discharge, slight adjustment of the manipulator might restore contact.

Initially positive deflections recorded from medial geniculate cells ranged from 0.5 to 25 mv in size. The transient 'injury potentials' were of much greater magnitude than this. The absolute size of the recorded potentials appar-
ently was related somehow to the electrode used, for frequently the voltage of all auditory units recorded with a particular electrode fell within narrow limits which appeared to be inversely related to electrode size. Partial spikes were noted on occasion with failure at about 30–50% of the spike height, a phenomenon already described for spinal motoneurons (7) and thalamic units (8).

**Types of Units Observed.** Table 1 summarizes the properties of the population of units studied. In all, about 322 units were seen. Of these, 44 were intensively studied, the remainder being seen only long enough to characterize the stimuli related to their firing. The first five categories in the table contain all the auditory units, though group five requires some clarification. The visual units met on the way to the medial geniculate were only briefly examined to verify their responsiveness to light and to approximate the electrode position. No unit was activated by both visual and auditory stimuli. This was in contrast to the reticular formation units described by Scheibel et al. (9) which were activated by both auditory and other stimulus modalities.

Many units proved to be unresponsive to all available stimuli. Most of these were located close to units that responded typically to acoustic stimuli. They often discharged spontaneously, sometimes intermittently, sometimes steadily, and slight movements of the electrode tip could evoke additional discharge. When a cell of this nature was silent for some time, the advancing microelectrode might evoke prolonged injury discharge or a short period of firing, followed by silence apparently due to dislodgment.

Over one-half of the auditory units encountered were activated by clicks. At or near threshold (5 clicks/sec.) the great majority fired only once or twice per click, and as intensity was progressively increased multiple firing became common, with as many as five or six spikes being evoked by clicks of moderate strength in extreme cases. With still further intensity increase, the number of discharges evoked might decline.

As for threshold determinations, a number of units were extremely sensitive, responding to clicks 20 db or more below the threshold for the $N_1$ response of the round window; most frequently stimuli 10–20 db above this value were required, and occasionally even more intense clicks than this were needed.

The above observations essentially confirm reports in the literature of similar studies on barbiturate-anesthetized cats. In addition, the latency of the auditory events recorded in the unanesthetized cat is also in good agreement with what has already been described (1, 2, 4). Slow waves were seen as early as 5 msec. and spikes as early as 6 or 7 msec. The great majority of units firing to clicks did so within 10 msec. or less, but some had much longer latencies, the extreme being 500 msec. Most units observed were studied for less than 5 minutes, but it was not uncommon to hold units for as long as 65 minutes.

**Response of Units to Repetitive Clicks at Various Frequencies.** In response to near-threshold clicks delivered at rates of 1–5/sec., medial geniculate units in the unanesthetized preparation respond 75–100% of the time. As the click frequency is increased they tend to fire less often, and ultimately their response may approach zero. These facts are illustrated for a representative unit in figures 2 and 3. For any given unit the relationship shown in figure 3 tends to be stable, and replications can be expected to yield nearly superimposable curves. As might be expected, higher response values will occur at a given click repetition rate when the stimulus intensity is raised.

The points on the curves in figure 3 were derived by counting the spikes occurring during the first 50 clicks of a much longer series. If only the first 25 responses are counted, slightly higher values are obtained at some points, while counting more than 50 responses changes the plotted values insignificantly. A sample size of 50, therefore, can be taken as a

<table>
<thead>
<tr>
<th>Unit Category</th>
<th>% of Total Observed</th>
<th>No. of Units</th>
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</thead>
<tbody>
<tr>
<td>1. Mainly click responsive</td>
<td>41</td>
<td>132</td>
</tr>
<tr>
<td>2. Mainly late responses to clicks</td>
<td>6</td>
<td>20</td>
</tr>
<tr>
<td>3. Mainly noise responsive</td>
<td>1</td>
<td>3</td>
</tr>
<tr>
<td>4. Mainly tone responsive</td>
<td>5</td>
<td>14</td>
</tr>
<tr>
<td>5. Not responsive to click, noise, tone, light, pinch or other</td>
<td>27</td>
<td>88</td>
</tr>
<tr>
<td>6. Light responsive</td>
<td>20</td>
<td>65</td>
</tr>
</tbody>
</table>
convenient and accurate measure of the capacity of a unit to follow repetitive clicks.

Of the 33 units studied in this manner, over 20 showed rather similar frequency response curves in that, when plotted, they clustered together and displayed a similar over-all slope. The click frequency at which response occurred 50% of the time ranged in this group from about 6/sec. to about 75/sec., with most occurring below 20/sec. In the remaining units the 50% point occurred at high click rates, in one case at 156/sec. and in another at over 200/sec.

Drug Effects. Limited success can be reported in attempts, with drugs, to modify the capacity of these units to respond to repetitive stimulation. Figure 4 illustrates the results obtained in the most successful of these attempts. Figure 4A shows the control frequency response curve of a unit like that shown in figures 2 and 3. After two determinations of this curve (with 50% points of 6.0 and 8.3 clicks/sec.), morphine (1 mg/kg) was injected intravenously; if this dose had any effect at all, it was to produce short-lasting diminution of excitability (50% points at 7.6 and 5.8 clicks/sec.). A second 1-mg/kg dose then was injected; an immediate increase in responsiveness was noted, and on the second and third determinations thereafter the 50% point rose to 18.3 and 17.6 clicks/sec., respectively (fig. 4B). A third dose of morphine was followed by a still greater elevation of the 50% points to 34.0 and 53.0 clicks/sec. (fig. 4C). A fourth dose then was injected, and this was quickly followed by high frequency discharge of the unit, and silence.

Two other effects, presumably due to the morphine, are worthy of mention, chiefly because they were dramatic and unexpected. The spontaneous activity of the units was increased well above that in the control and, following a click the units tended to respond repetitively.

DISCUSSION

The method of testing excitability of neural units by recording their successive responses to equally spaced stimuli, while not novel, appears to be a valuable and practical tool for the study of auditory units. With a few exceptions, if a unit responds at all to clicks, relations like those of figure 3 can be described for it. The curves are stable and can be repeated within reasonably close limits. Preliminary studies show they can be modified deliberately by changing the physiological state of the animal.

On the general question of diminution of response at higher frequencies of stimulation,
evidence is available from studies upon several functional neuronal groups (7-10). Mountcastle and Rose (11) classify tactile thalamic cells as either 'adapting units' or 'cut-off units,' depending upon their behavior toward repetitive stimulation. The first group responds to each stimulus up to frequencies around 20-100/sec., at higher stimulus frequencies a constant maximal rate of discharge is evoked. In contrast, 'cut-off units' respond one-for-one up to 5-20 stimuli/sec., above which their response abruptly drops to zero; stimulus frequencies higher than this evoke little response or none at all. With very few exceptions, the medial geniculate units reported here are of the 'cut-off' type. It must be recalled, however, that only weak stimuli have been employed in this study, and perhaps 'adapting units' would be found in numbers if other and more intense sounds were used.

From one medial geniculate unit to the next a considerable range exists in the frequency of stimulation at which firing dropped to 50% of the maximum. The factors that determine this 50% point are not well understood. It could be extended undoubtedly below the limit indicated in the present study, if units firing repetitively with long latencies were included; for reasons of time economy this was not done. These 50% points were also shifted when intensity of stimulation was either decreased or increased as compared to the standard value used in this study, but these relations were not explored extensively. Finally, it was the general impression that units observed in the unanesthetized preparation may have been more sensitive to excitation by very weak stimuli than previously has been reported from studies under barbiturates.

On the matter of drugs and their effect upon excitability of units, the data presented here, while somewhat meager, show morphine to produce a trend toward increased firing. Thus, 'spontaneous' activity increased, repetitive firing appeared, and a rise in the 50% point occurred. This rise in responsiveness may be related to the fact that most cats, when given morphine (2-5 mg/kg), show increased startle response and other evidence of hyperactivity (12).

The expert assistance of Mr. Allen Rupert in the performance of these experiments is gratefully acknowledged.

REFERENCES