The Journal of Neuroscience, February 1, 1999, *19*(3):1149–1163

Interaural Intensity Difference Processing in Auditory Midbrain Neurons: Effects of a Transient Early Inhibitory Input

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Interaural intensity differences (IIDs) are important cues that animals use to localize high-frequency sounds. Neurons sensitive to IIDs are excited by stimulation of one ear and inhibited by stimulation of the other ear, such that the response magnitude of the cell depends on the relative strengths of the two inputs, which in turn depends on the sound intensities at the ears. In the auditory midbrain nucleus, the inferior colliculus (IC), many IID-sensitive neurons have response functions that decline steeply from maximum to zero spikes as a function of IID. However, there are also many neurons with much more shallow response functions that do not decline to zero spikes. We present evidence from single-unit recordings in the Free-tailed bat's IC that this partially inhibited response pattern is a result of the inhibitory input to these cells being very brief (~2 msec). Of the cells sampled, 54 of 137 (40%) achieved partial inhibition when tested with 60 msec tones, and the inhibition to these 54 cells occurred primarily during the first few milliseconds of the excitatory response. Consequently, the initial component of the response was highly sensitive to IIDs, whereas the later component was primarily insensitive to IIDs. Each of the 54 "partially inhibited" cells was able to reach complete inhibition with very short stimuli, such as simulated bat echolocation calls that invoked only the initial, IID-sensitive component. Local application of inhibitory transmitter antagonists disabled the short inhibitory input, indicating that this response pattern is created within the IC.

Key words: interaural intensity difference; inferior colliculus; sound localization; inhibition; auditory processing; binaural

Insectivorous bats are extremely good at localizing sounds in space. Bats, like other mammals, use binaural cues such as interaural intensity differences (IIDs) to accomplish this task (Erulkar, 1972; Mills, 1972; Irvine, 1986). The basic neural circuitry associated with processing IIDs is superficially straightforward: neurons that are sensitive to IIDs are excited by stimulation of one ear and inhibited by stimulation of the other ear. Thus, a given IID generates a combination of excitation and inhibition that is reflected in a cell's spike count. In general, when a sound has an IID that is more intense at the excitatory ear, it generates a higher spike count than when the same sound has an IID that is more intense at the inhibitory ear.

A substantial population of IID-sensitive neurons is found in the midbrain auditory center, the inferior colliculus (IC) (Pollak et al., 1986; Semple and Kitzes, 1987; Irvine and Gago, 1990; Park, 1998). Many of these cells have IID functions that decline sharply from maximum to zero spikes as IIDs are varied. Hence, these cells appear to be well suited to code IIDs in that the same sound coming from different locations in space elicits different spike counts. However, other collicular cells, approximately one-third to one-half of the IID-sensitive IC cells, have response functions that are relatively shallow and do not go to zero spikes (Wenstrup et al., 1988; Irvine and Gago, 1990; Park and Pollak, 1993; Park, 1998). Compared to the cells with sharp IID functions that go to zero spikes, these cells achieve only partial inhibition and thus in terms of spike counts appear to be less well suited for coding IIDs. Both the underlying mechanism(s) that results in partial versus complete inhibition and the possible functional significance, if any, of the partially inhibited response type have remained unanswered questions for some time. The present study was designed to elucidate these questions.

Using standard pure tone stimuli, we found that the partially inhibited response type was primarily shaped, within the IC, by a transient inhibitory input acting on the first few milliseconds of the spike train. We also found that when we presented the same cells with transient stimuli, their IID functions dramatically changed into completely inhibited functions. Hence, the IID sensitivity of these cells was highly dependent on stimulus duration.

MATERIALS AND METHODS

Surgical and recording procedures. Eleven Mexican free-tailed bats, Tadarida brasiliensis mexicana, were experimental subjects. The experimental protocol was approved by the University of Illinois at Chicago institutional animal care and use committee. Before surgery, animals were anesthetized with methoxyflurane inhalation and 15 mg/kg sodium pentobarbital injected subcutaneously. The hair on the bat's head was removed with a depilatory, and the head was secured in a head holder with a bite bar. The muscles and skin overlying the skull were reflected, and 4% lidocaine hydrochloride was applied topically to the open wound. The surface of the skull was cleared of tissue and a ground electrode was placed just beneath the skull over the posterior cerebellum. A layer of small glass beads and dental acrylic was placed on the surface of the skull to secure the ground electrode and to serve as a foundation layer to be used later for securing a metal rod to the bat's head.

The bat was transferred to a heated (27–30°C), sound-attenuated room, where it was placed in a restraining apparatus attached to a custom-made stereotaxic instrument (Schuller et al., 1986). A small metal

Received May 29, 1998; revised Nov. 4, 1998; accepted Nov. 20, 1998.

This work was supported by National Institutes of Health Grant DC02850 and The Biological Resource Laboratory, University of Illinois at Chicago.

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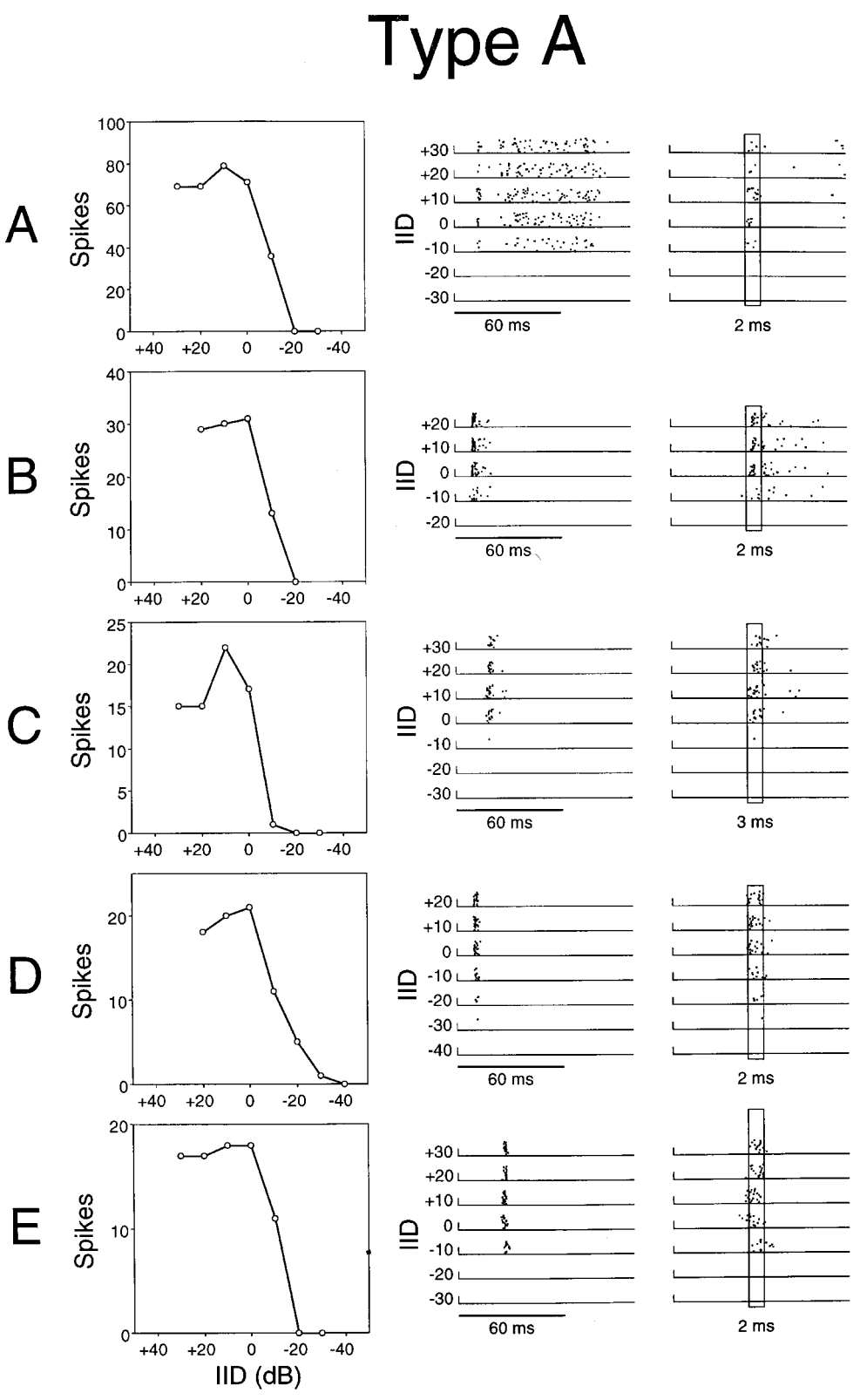

Figure 1. Examples of IID-sensitive neurons recorded from the inferior colliculus. For each cell, an interaural intensity difference (*IID*) function is plotted on the *left*, showing spike count as a function of IID. A corresponding raster plot, displaying spike activity for 100 msec beginning at stimulus onset, is plotted in the *center*. An expanded raster plot, focusing on the beginning of the spike train, is shown on the *right* (the time scale for each set of expanded rasters was selected to best show the effects of IID on the initial spikes). *A–E*, IID functions and raster plots from cells with spike counts that reached complete inhibition with increasing intensities to the ipsilateral, inhibitory ear (negative IIDs). For convenience, we refer to these cells as Type A cells. *F–J*, IID functions and raster plots from cells that only reached partial inhibition with increasing intensities to the inhibitory ear. For convenience, we refer to these cells as Type B cells. Stimuli were 60 msec tones presented at each cell's characteristic frequency. Note that Type B cells were characterized by an early inhibitory component for IIDs favoring the inhibitory ear, emphasized by the *boxes* superimposed on the expanded rasters. *(Figure continues.)*

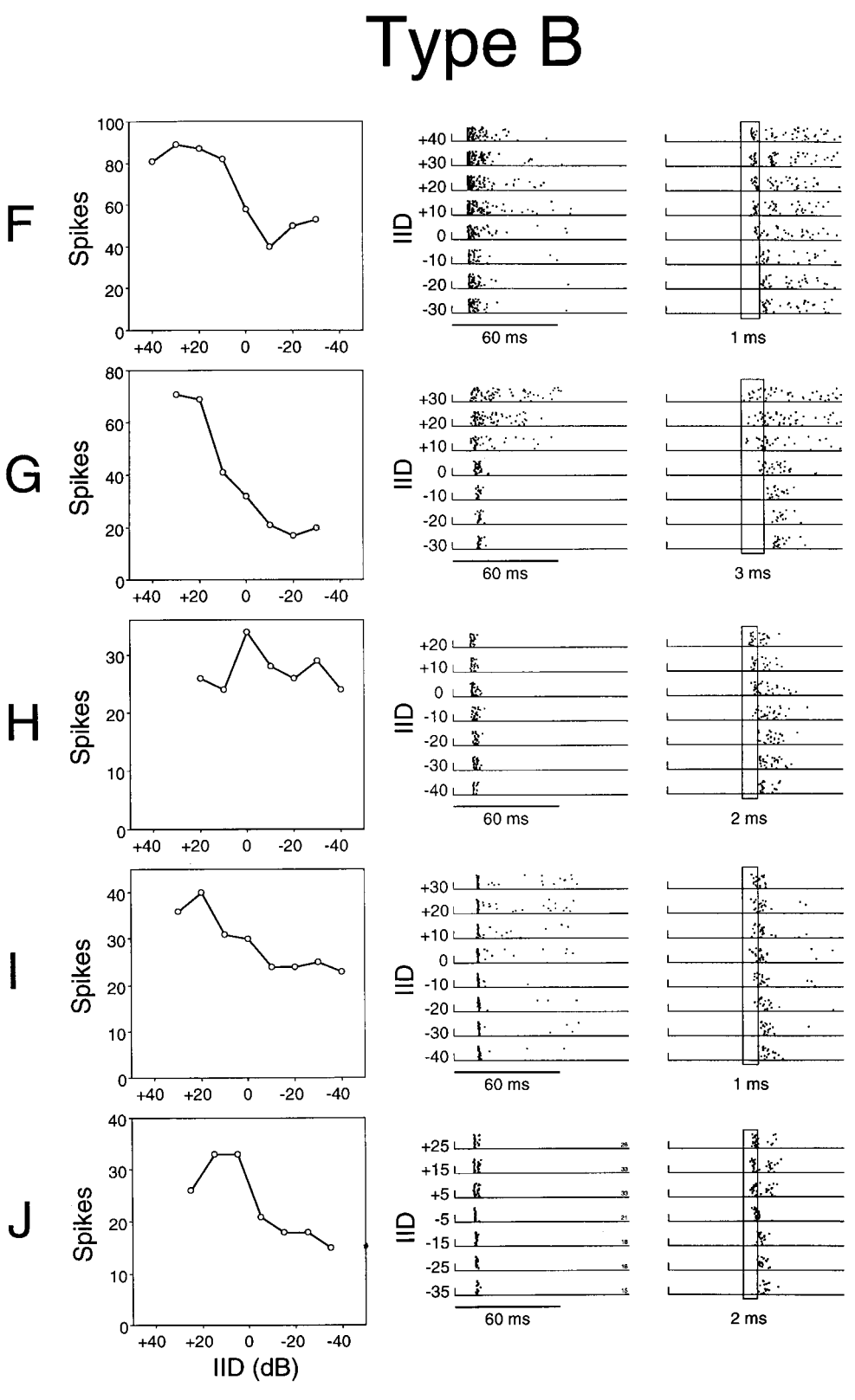

Figure 1 continued.

rod was cemented to the foundation layer on the skull and then attached to a bar mounted on the stereotaxic instrument to ensure uniform positioning of the head. A small hole (~0.5–1.0 mm diameter) was then cut over the inferior colliculus on one side. The position of the hole and positioning of the electrode to reach the IC followed procedures described by Schuller et al. (1986). Recordings were begun after the bat was awake. The local anesthetic was refreshed every 30 min. If an animal became agitated and began moving around during the course of a recording session, an additional subanesthetic injection of sodium pentobarbital (10 mg/kg body weight) was given subcutaneously. This dosage

of pentobarbital never induced anesthesia. The bats were still awake: their eyes were open, they often drank water when it was offered, and they responded when their face or ears were gently touched. There were no noticeable, systematic changes in neuronal response properties from the subanesthetic dose of pentobarbital. Additional pentobarbital injections were administered on only a few occasions and then only once during a given recording session. Recording sessions generally lasted from 3 to 5 hr per day to minimize the animals' discomfort from being restrained.

Action potentials were recorded with a glass pipette filled with buffered 1 M NaCl, and electrode impedance ranged from 5 to 20 MΩ. Electrode penetrations were made vertically through the exposed dorsal surface of the inferior colliculus. Subsequently, the electrode was advanced from outside of the experimental chamber with a piezoelectric microdrive.

Acoustic stimuli and data acquisition. When a unit was encountered, we first determined its characteristic frequency and absolute threshold audiovisually to set stimulus parameters subsequently controlled by computer. The characteristic frequency was defined as the frequency that elicited responses at the lowest sound intensity to which the unit was sensitive. Binaural stimuli were then presented to determine whether the unit was monaural or binaural, and if it was binaural, whether it was primarily excited by sound at each ear or whether it was primarily excited by one ear and primarily inhibited by the other ear, which is the pattern customarily associated with IID sensitivity.

Each cell was tested with 60 msec tones and 2 or 4 msec linear frequency sweeps. Stimuli were presented at a rate of four per second. Tone stimuli were presented at a cell's characteristic frequency. The sweep stimuli swept downward from 5 kHz above to 5 kHz below a unit's characteristic frequency. The sweeps at both ears were coherent in that they had the same frequency range and duration, and each began and ended with the same phase. Additional stimuli used with some cells included 2 msec tones at the cell's characteristic frequency, and 60 or 100 msec tones with sinusoidal modulations in amplitude. Amplitude-modulated tones were presented at the characteristic frequency, with a modulation depth of 100%. All stimuli had 0.2 msec rise and fall times shaped by a sigmoid. Stimuli were presented via Brüel and Kjaer ¼ inch microphones used as ear phones fitted with probe tubes (5 mm diameter) that were placed in the funnel of each pinna. Maximum sound intensity was 90 dB sound pressure level (SPL) measured 0.5 cm from the opening of the probe tubes. Sound pressure and the frequency response of each earphone was measured with a ¼ inch Brüel and Kjaer microphone. Each earphone showed less than ±8 dB variability for the frequency range usually used (15–80 kHz), and intensities between the earphones did not vary more than ±3 dB at any of those frequencies. For fundamentals between 10 and 50 kHz and 74 dB SPL (the highest intensity we usually used), all harmonics were at least 35 dB less intense than the fundamental. For higher frequencies, the harmonics were even lower.

A power spectrum analysis was performed to assess the spectral content of the very short stimuli. For the 2 msec sweeps, the intensity of frequencies outside the range of the sweep was ~40 dB or more below the intensity of the sweep frequencies. For the 2 msec tones, energy peaked at the tone frequency: at 20 dB below the peak the bandwidth of spectral splatter was ~5 kHz and at 40 dB below the peak the bandwidth was ~10 kHz.

Acoustic isolation between the ears was better than 40 dB, determined empirically during the course of the experiments by testing units that were operationally defined as monaural: units that were excited by stimulation of the excitatory, contralateral ear, but showed no apparent excitatory or inhibitory effects when the ipsilateral ear was stimulated. To determine aurality, the intensity at the contralateral ear was set 20 dB above the cell's threshold at its characteristic frequency, and the ipsilateral ear was stimulated with intensities ranging from ~40 dB below to 40 dB above that at the contralateral ear. Next, to determine acoustic isolation, the contralateral stimulus was turned off and the intensity at the ipsilateral ear was increased until spikes were evoked, presumably via cross-talk to the contralateral ear.

IID functions were generated by holding the intensity at the contralateral, excitatory ear constant at ~20 dB above a unit's threshold and varying the intensity at the ipsilateral, inhibitory ear in 10 dB steps ranging from ~40 dB below to 40 dB above the intensity at the excitatory ear. Twenty stimulus repetitions were presented at each IID tested, and the order of presentation was varied pseudorandomly.

Twelve cells were tested before and during microiontophoretic application of the inhibitory transmitter antagonists bicuculline and strychnine, following established procedures (Havey and Caspary, 1980; Park and Pollak, 1993). The antagonists block postsynaptic receptors for inhibitory transmitters and thus prevent the transmitters from binding and exerting their effects. We used a combination of bicuculline and strychnine to block the primary inhibitory transmitters of the auditory midbrain, GABA and glycine, respectively (Klug et al., 1995; Winer et al., 1995).

RESULTS

This study reports on 137 IID-sensitive neurons recorded from the IC of the Mexican free-tailed bat. Each cell in our sample showed a prominent excitation for sound presented to the contralateral ear and a prominent inhibition for sound presented to the ipsilateral ear. For convenience, we will hereafter refer to the ear that evoked excitation as the excitatory ear and the ear that evoked inhibition as the inhibitory ear. We also encountered, but did not include in our analysis, 24 monaural cells, 21 cells that received prominent excitation from both ears, and 7 cells that did not respond to our sweep stimuli.

In the following sections we will first describe how IID-sensitive cells responded to IIDs within the biologically relevant range that the animal's head would generate in the free field (approximately ±30 to 40 dB) (Harnischfeger et al., 1985). Specifically, we will focus on the degree to which spikes could be suppressed by sound presented to the inhibitory ear. We will then focus on the cells in our sample (40%) that did not achieve complete spike suppression at any biologically relevant IID when presented with 60 msec tone stimuli. We will show how the time course of the inhibitory input to these cells was fundamentally different from that of cells that achieved complete inhibition. Finally we will describe how this large subpopulation of IC cells responded to IIDs in a strikingly different way when much shorter stimuli were presented and how iontophoretic application of neurotransmitter antagonists affected their responses.

IID-sensitive cells could be categorized into two groups based on the degree of inhibition they achieved with 60 msec tones

The goal of our study was to explore IID-sensitive cells in the inferior colliculus that cannot be completely inhibited by any IID within the biologically relevant range when standard, pure tone stimuli are used. Hence, we first assessed IID sensitivity among our sample to identify those cells. With 60 msec tones, we found that 60% (83 of 137) of the IID-sensitive cells achieved complete inhibition as the intensity to the inhibitory ear was increased, whereas 40% (54 of 137) achieved only partial inhibition. Figure 1 shows the IID functions and corresponding raster plots of cells in both categories. Figure 1*A–E* shows data from five cells that achieved complete spike suppression. For convenience, we hereafter refer to cells that behaved in this way as Type A cells. Figure 1*F–J* shows data from five cells that achieved only a partial spike suppression. For convenience, we hereafter refer to cells that behaved in this way as Type B cells.

Among the Type B cells, the percentage inhibition attained as IIDs changed from favoring the excitatory ear to favoring the inhibitory ear varied from cell to cell. Percentage inhibition was calculated by taking the percentage decrease in spike count from the maximum number of spikes to the minimum number of spikes on a cell's IID function. The histogram in Figure 2 shows the distribution for percentage inhibition for all of the cells that we tested. Type B cells are indicated by the hatched bars. As the histogram shows, some cells displayed only a minor decrease in

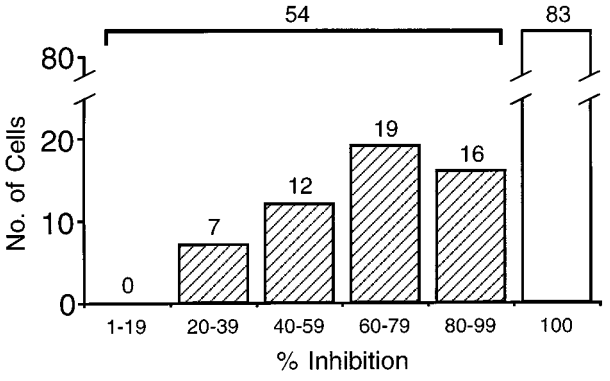

Figure 2. Distribution of percentage inhibition for the 137 cells studied. Percentage inhibition was calculated by taking the percentage decrease in spike count from the maximum number of spikes to the minimum number of spikes on a cell's IID function. The 54 partially inhibited Type B cells are indicated by the *hatched bars*. The 83 completely inhibited Type A cells are represented by the *open bar* at 100% inhibition.

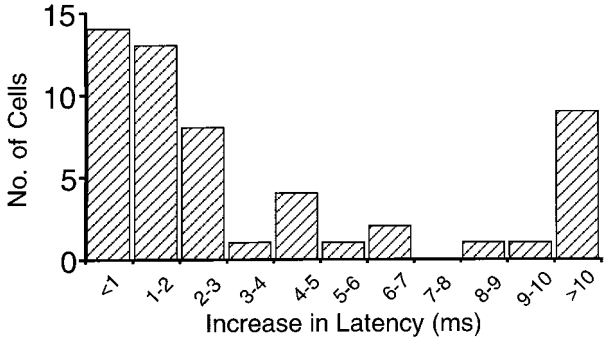

Figure 3. Durations of the early, inhibitory component of Type B neurons. Duration of inhibition was estimated from the change in response latency associated with changing IIDs. The median first spike latency was calculated for each IID for each cell. Then, to generate the distribution shown here, the latency value of the most positive IID was subtracted from the latency value of the most negative IIDs.

spike count of 20–39%, whereas others displayed a substantial decrease of 80–99%. The 83 cells that achieved complete inhibition are represented by the open bar at 100%. By including the completely inhibited (Type A) cells, the distribution shown in this histogram might suggest a continuum for degree of inhibition among the population of IC cells from slight to moderate to complete inhibition. However, the further analyses that we present below show that Type A and Type B cells are indeed distinct in that the time course of inhibition is fundamentally different for the two response types.

Type B (partially inhibited) IC cells were characterized by an early, short duration inhibition from the inhibitory ear

We found that each of the Type B cells (54/54, 100%) shared a common, defining response feature. Each cell was strongly inhibited during the first few milliseconds of the spike train for IIDs favoring the inhibitory ear. This feature was readily observable when the time base of the cells' raster plots was expanded (Fig. 1*F–J*, expanded rasters). In Figure 1 (and later figures) the raster plots displayed on the right replot the initial portions of the rasters on the left, but on an expanded time scale. For example, the expanded raster plots for the cell in Figure 1*F* show that the initial spikes observed for IIDs favoring the excitatory ear (+40 to 0 dB) were inhibited for IIDs favoring the inhibitory ear (−20 and −30 dB). In other words, increasing the intensity at the inhibitory ear completely suppressed the early spike activity (emphasized in Fig. 1*F* by the box corresponding to 2 msec). The same pattern can be seen for each of the Type B cells in Figure 1.

Some of the Type B cells in our sample also showed signs of later inhibitory components that affected later spike activity (e.g., Fig. 1*G*). Later inhibitory components could only be documented for cells that expressed sustained spike activity evoked by the excitatory ear. Of the 54 Type B cells in our sample, 22 cells had this type of sustained activity, whereas the remaining 32 cells responded only to the onset of stimulation to the excitatory ear. We found that within the subpopulation of Type B cells that had a sustained response pattern, more than half (14 of 22) exhibited later inhibitory components that suppressed later spike counts by 50% or more (Fig. 1*G,I*). Nevertheless, we reemphasize that each of the 54 partially inhibited (Type B) cells showed a strong, short-lasting inhibitory component at the very beginning of the spike train that distinguished them from Type A cells.

The early, short duration inhibition described above resulted in two important effects. First, the inhibition of early spikes associated with increasing intensities at the inhibitory ear resulted in partial inhibition of the cell's overall spike count. This effect can be seen in the expanded raster plots for each of the Type B cells in Figure 1. In contrast, for each of the Type A cells in Figure 1, the inhibition had a duration that was sufficient to completely overlap the spike train, causing the IID functions of these cells to achieve 100% inhibition. Second, the early inhibition observed for the partially inhibited cells caused the response latency for each of these cells to increase as IIDs changed from favoring the excitatory ear to favoring the inhibitory ear. This effect can also be seen in the expanded raster plots for the Type B cells in Figure 1, emphasized by the boxes drawn around the initial component of the spike trains.

The change in response latency of Type B cells associated with increasing intensities to the inhibitory ear was used to estimate the duration of the initial short inhibitory component. For example, the cell shown in Figure 1*F* had a median first spike latency of 9.10 msec when the tone was presented at an IID of +40 dB (40 dB more intense at the excitatory ear). When the intensity to the inhibitory ear was increased to generate an IID of −30 dB, the latency increased to 9.77 msec. Hence, for this cell, we estimated the duration of the initial inhibitory component to be 0.67 msec. For the cell shown in Figure 1*G*, the response latency increased by 2.50 msec. Note that this method of estimating the duration of inhibition only applied to inhibition that was large enough to completely cancel the excitation. Also note that the duration of inhibition increased as a function of the intensity at the inhibitory ear (as seen in the expanded rasters for Fig. 1*F,G*). This type of intensity-dependent increase in the duration of inhibition has been reported previously (Irvine et al., 1995; Park, 1998). Our estimates were always based on the highest intensity available to the inhibitory ear (the most negative IID tested for each cell).

The distribution of inhibitory durations for the 54 Type B cells is presented in Figure 3. The majority of cells had inhibitory durations between 0.5 and 5.0 msec, and the median value was 2.03 msec. However, as the distribution in Figure 3 shows, the initial inhibition for a number of cells had relatively long durations. The raster plots for the cells with the largest values revealed that they responded to stimulation of the excitatory ear with one or a few initial spikes followed by a period of silence and then more spike activity. This temporal response pattern is seen for

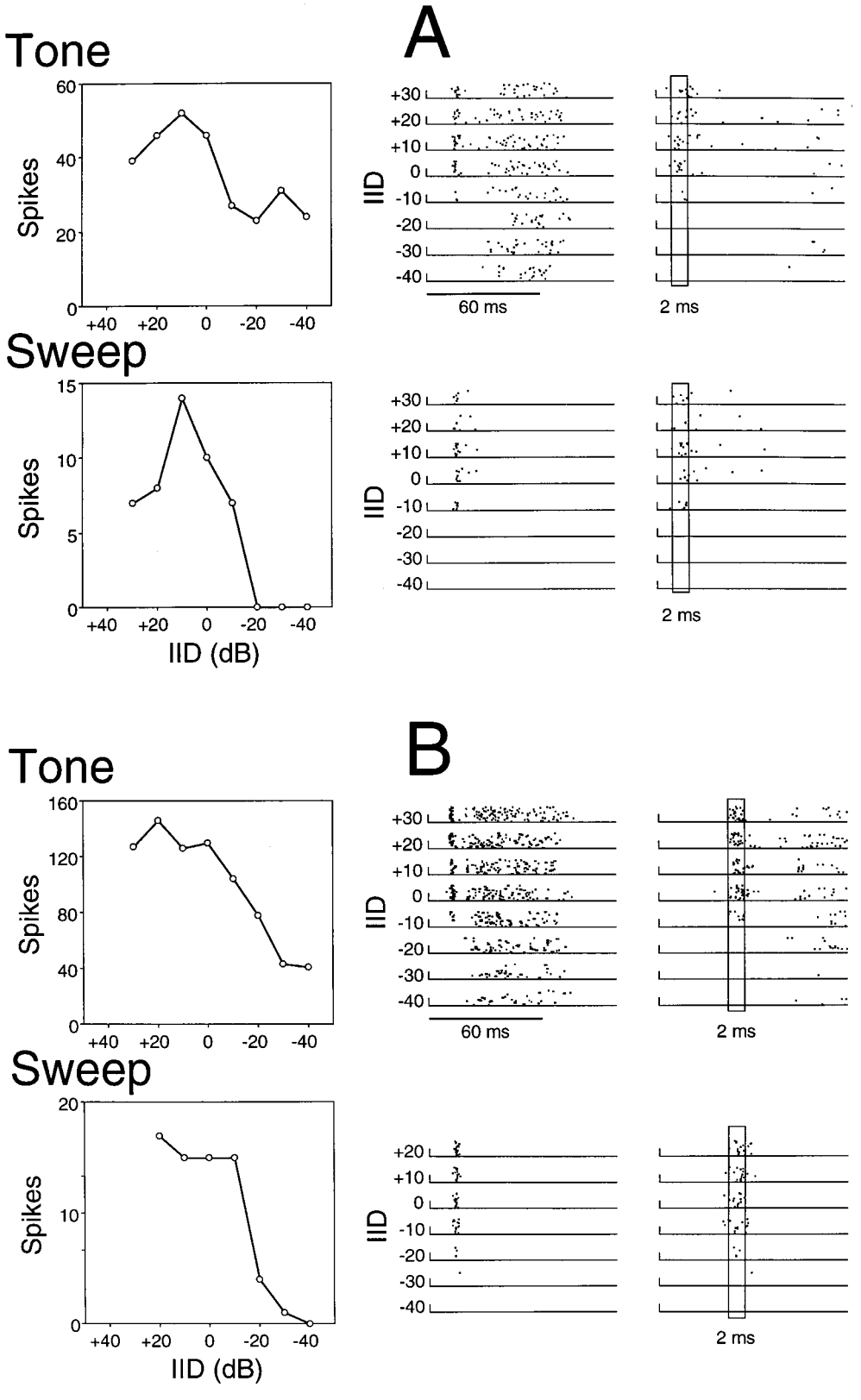

Figure 4. IID functions and raster plots for two Type B cells that responded to stimulation of the excitatory ear with one or a few initial spikes followed by a period of silence and then more spike activity (primary with notch response type). The general format follows that of Figure 1. The *top panel* for each cell, labeled *Tone*, shows responses to 60 msec tones. For both cells, the period of silence in the excitatory response appears to have exaggerated the effect of the early inhibitory component evoked by the inhibitory ear. The *bottom panel* for each cell, labeled *Sweep*, shows how these cells responded to short sweep stimuli. As for each of the Type B cells studied, the IID functions of these two Type B cells reached complete inhibition with short sweeps. Note that here and in later figures the time scale for the raster plots on the *left* is the same for tones and sweeps.

the raster plots generated by positive IIDs that favored the excitatory ear, and similarly for stimulation of the excitatory ear alone (not shown). Figure 4 presents the IID functions and raster plots for two such cells that appeared to have long initial inhibitory components (Fig. 4*A,B, top panels*). For both of these cells, the initial inhibitory component, driven by the inhibitory ear, suppressed the initial spikes, but because of the silent period that normally followed the initial spikes, it did not necessarily last as long as the change in latency might suggest (32.6 and 25.05 msec for the cells in Fig. 4, *A* and *B*, respectively). In other words, the

temporal response pattern of these cells, generated by the excitatory ear, may have exaggerated the apparent duration of the initial inhibitory component. The bottom panels in Figure 4*A,B* show how these cells responded to short-frequency sweeps, the topic of the next section.

The IID functions of Type B cells changed dramatically to resemble those of Type A cells when shorter stimuli were used

The short time course of inhibition that we observed for the partially inhibited cells led us to hypothesize that a very brief stimulus might make it possible for these cells to reach complete inhibition. The rationale for this hypothesis was that a very short stimulus could evoke an excitation and an inhibition with equally short durations. If the short excitation and inhibition completely overlapped, then spikes should be completely inhibited for IIDs favoring the inhibitory ear. If this scenario were true, then the IID functions of Type B cells would change dramatically depending on the stimulus duration: they would be only partially inhibited for tones longer than a few milliseconds but completely inhibited for shorter stimuli.

To assess the above hypothesis, we tested each of the 54 Type B cells with short frequency sweeps descending from 5 kHz above to 5 kHz below a cell's best frequency. For the majority of the cells (47), we used 2-msec-long sweeps. We used 4- or 5-msec-long sweeps for the remaining seven cells because those cells did not respond to 2 msec sweeps. Short-frequency sweeps were selected for two reasons. First, we had previously used this type of stimuli in earlier studies and found that they reliably evoked a very punctate excitation and inhibition (Park et al., 1996; Park, 1998). Second, in many respects, these frequency sweeps simulate the free-tailed bat's echolocation calls and therefore have the added feature of being biologically relevant signals.

As predicted, each of the 54 cells that were only partially inhibited with 60 msec tones were completely inhibited with short frequency sweeps. The IID functions and raster plots for four Type B cells are shown in Figure 5. For each of these cells, the top panel shows responses to 60 msec tones, whereas the bottom panel shows responses to short sweep stimuli. In every case, the IID function went to zero spikes with the short sweeps. Similar sweep data are also presented for the cells in Figures 4, 7, and 9.

We also tested sweep stimuli on 55 of the Type A cells. Of the 55 Type A cells, 54 had IID functions that reached complete inhibition both for 60 msec tones and for sweeps, and two examples are shown in Figure 6.

One reason that we initially selected sweeps as a stimulus was that sweeps elicit very short periods of excitation and inhibition. However, because sweeps cross many frequency channels, the data collected with sweep stimuli might reflect excitatory and inhibitory inputs tuned to different frequencies. This raises the possibility that frequency tuning, not stimulus duration, might account for the differences in IID functions between 60 msec tones and sweeps. To further test our hypothesis that stimulus duration determines the degree of inhibition for Type B cells, we tested 11 Type B cells with 2 msec tones. The results showed that, as with the sweeps, the IID function of each cell tested reached complete inhibition with very brief tone stimuli. The data from two Type B cells tested with 2 msec tones are presented in Figure 7. As Figure 7 shows, the IID functions of Type B cells were very similar for 2 msec tones and short sweeps, indicating that stimulus duration does indeed appear to play a key role in determining how these cells respond to IIDs.

The results presented above led us to try yet another stimulus variation on some of the Type B cells: sinusoidally amplitude-modulated (SAM) tones. SAM stimuli are commonly used to assess a neuron's response to amplitude modulations, one of the most important features common to communication, and other naturally occurring sounds (Langner, 1992; Muller-Preuss et al., 1994; Saberi and Hafter, 1995; Grothe et al., 1997). Our hypothesis concerning stimulus duration would predict that Type B cells might respond differently to low and high modulation rates of SAM stimuli. The rationale of this idea was similar to the rationale that led us to use sweeps and 2 msec tones. In essence, each cycle of a periodic stimulus can be conceptualized as an individual stimulus event in which each cycle of a low modulation rate has a longer duration than each cycle of a higher rate (Fig. 8, *top*). Hence, we predicted that Type B cells might generate partially inhibited IID functions for low modulation rates and completely inhibited IID functions for high modulation rates.

We tested 10 Type B cells using SAM signals with modulation rates ranging from 50 to 500 Hz. The IID functions for 9 of the 10 cells conformed with our prediction in that they had partially inhibited IID functions for low modulation rates and completely inhibited IID functions for higher modulation rates. Three examples are shown in Figure 8.

Blocking of inhibitory inputs indicated that the Type B response pattern is shaped within the IC

To determine whether the Type B response pattern is an emergent response property of the IC or whether it reflects an integration accomplished below the IC, we experimentally blocked the inhibitory inputs to Type B cells while recording their responses to binaural sweep stimuli. We blocked inhibitory inputs by simultaneously microiontophoresing the inhibitory transmitter antagonists bicuculline and strychnine onto individual IC cells.

In each of the 12 Type B neurons that we tested in this manner, the antagonists greatly reduced the short-lasting inhibition that characterized the Type B response pattern. Thus, the initial component of the response was no longer completely inhibited during application of the antagonists, supporting the hypothesis that this response type is an emergent property of the IC. Figure 9 shows two examples of Type B cells tested before application of the antagonists and during application. Before application, the cells showed the Type B response pattern with 60 msec tones and short sweeps. The early inhibitory component suppressed early spikes, generating a partially inhibited IID function with the tone and a completely inhibited IID function with the sweep. However, when inhibitory inputs were locally blocked, the effectiveness of the inhibitory component was substantially reduced, changing the shape of the IID functions generated with sweeps. The fact that these functions still retained a substantial inhibition even after inhibitory transmitters were blocked suggests that, in addition to integrating inhibitory inputs within the IC, the cells' response did reflect, to some extent, additional binaural interactions at a lower level.

DISCUSSION

Similar to previous reports, we observed two types of IID-sensitive cells in the inferior colliculus: cells that had steep IIDfunctions and were completely inhibited at certain IIDs, and cells that had IID functions with relatively shallow slopes that did not decline to complete inhibition (Wenstrup et al.,

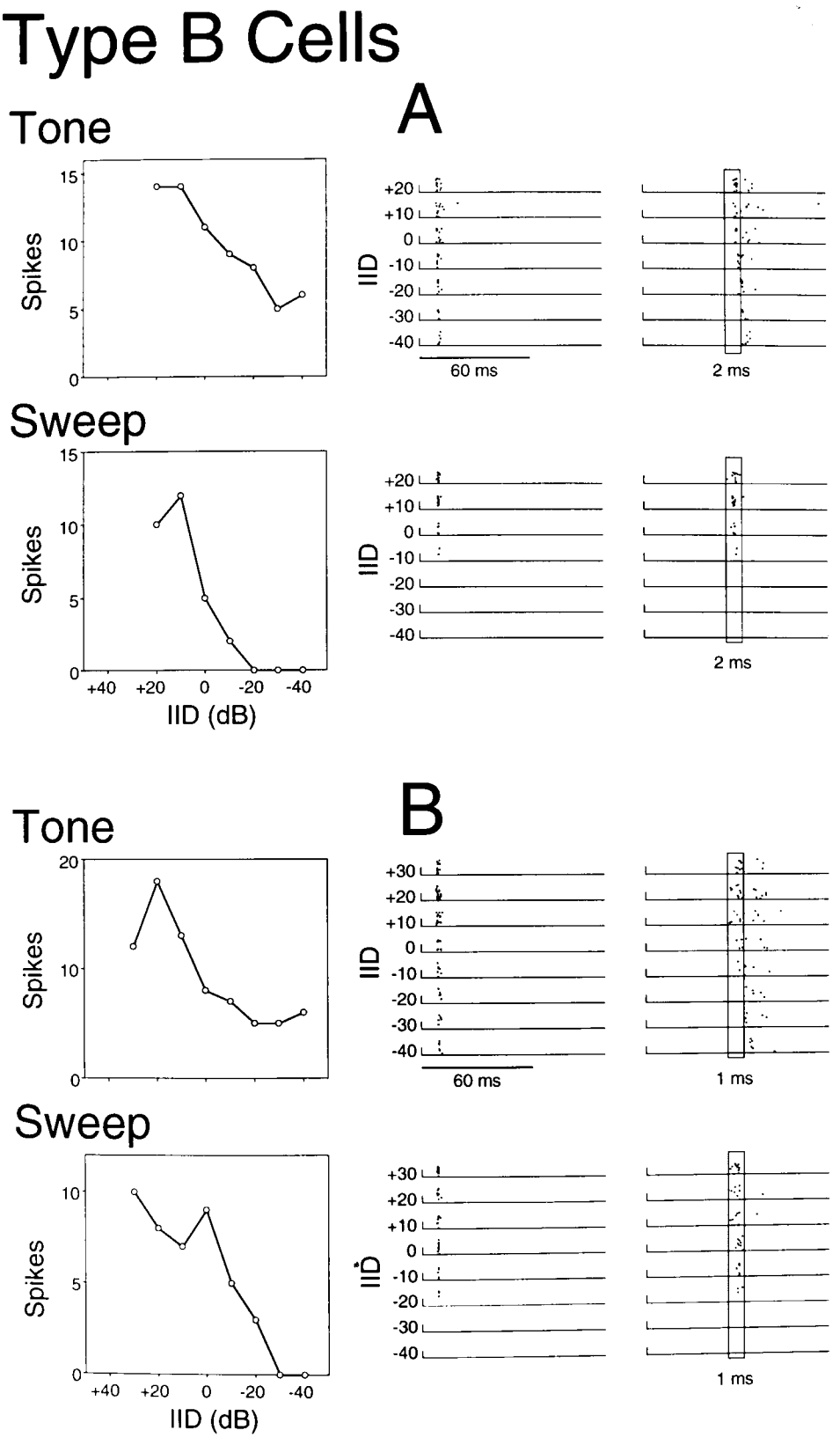

Figure 5. Four examples of Type B cells tested with 60 msec tones (*top panels*) and short frequency sweeps (*bottom panels*). The general format follows that of Figure 1. As for each of the partially inhibited Type B cells studied, these cells became completely inhibited with sweep stimuli. *(Figure continues.)*

1988; Irvine and Gago, 1990; Park and Pollak, 1993; Park, 1998). The new finding presented in this report is that each of the Type B cells received a strong but transient ipsilaterally driven inhibition that acted on the first few milliseconds of contralaterally evoked excitation. As a result, the first few milliseconds of each Type B cell's response was sharply sensitive to IIDs, such that the initial spikes became completely inhibited by IIDs that were more intense at the ipsilateral ear than the contralateral ear. In contrast, spikes occurring after the first few milliseconds were never completely inhibited.

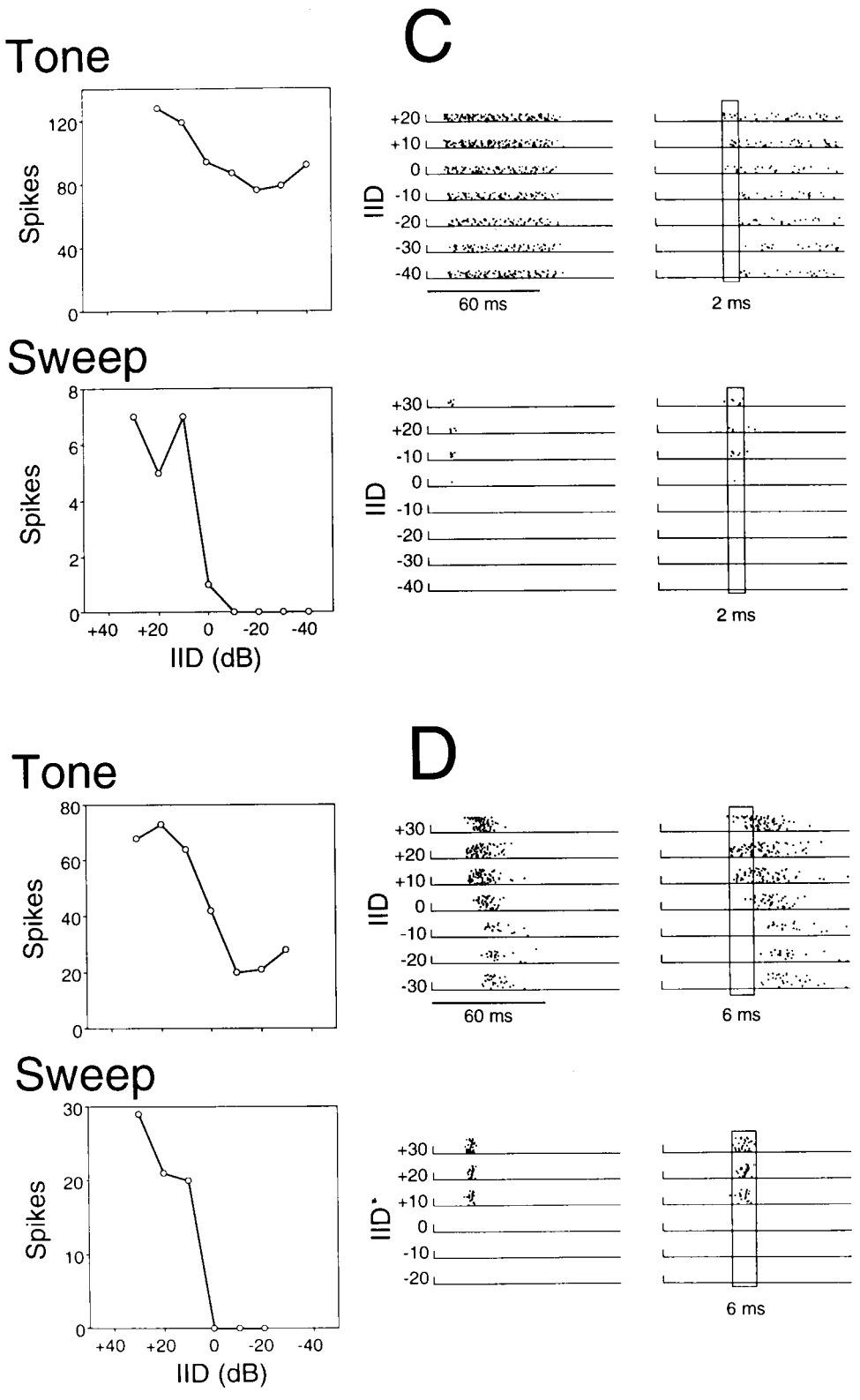

Figure 5 continued.

IID sensitivity of Type B cells is dependent on stimulus duration

The transient, ipsilateral inhibition to Type B cells made the first few milliseconds of the cells' response sharply sensitive to a sound's IID. This finding was consistent for each stimulus we tested: 60 msec tones, 2 msec tones, short-frequency sweeps, and amplitude-modulated tones. For short stimuli (2 msec tones, short sweeps, and SAMs with short cycle durations), the contralaterally

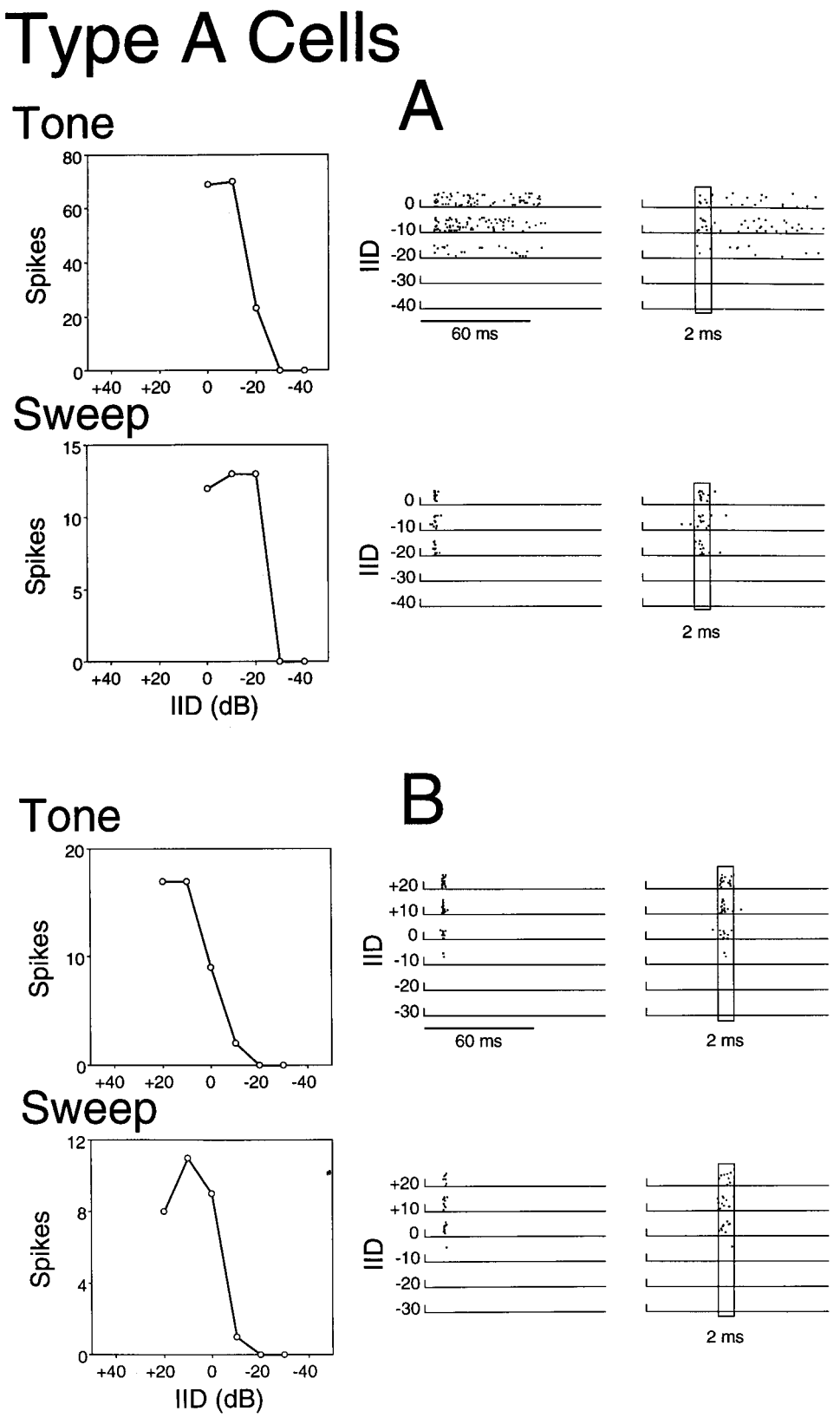

Figure 6. Two examples of Type A cells tested with 60 msec tones and short frequency sweeps. The general format follows that of Figure 1. Note that the IID functions of Type A cells reach complete inhibition for both stimuli.

evoked excitation overlapped in time with the ipsilaterally evoked early inhibition, and thus, for short stimuli, Type B cells behaved like Type A cells: their IID functions declined steeply to zero spikes as a function of IID. For long stimuli (60 msec tones and SAMs with long cycle durations), the first few milliseconds of spike activity also declined steeply to zero spikes as a function of IID. However, the later part of the cell's response was unaffected by the early transient inhibition, such that some or all later spikes persisted. Thus, for longer stimuli, Type B cells were never completely inhibited. Hence, the degree of binaural inhibition

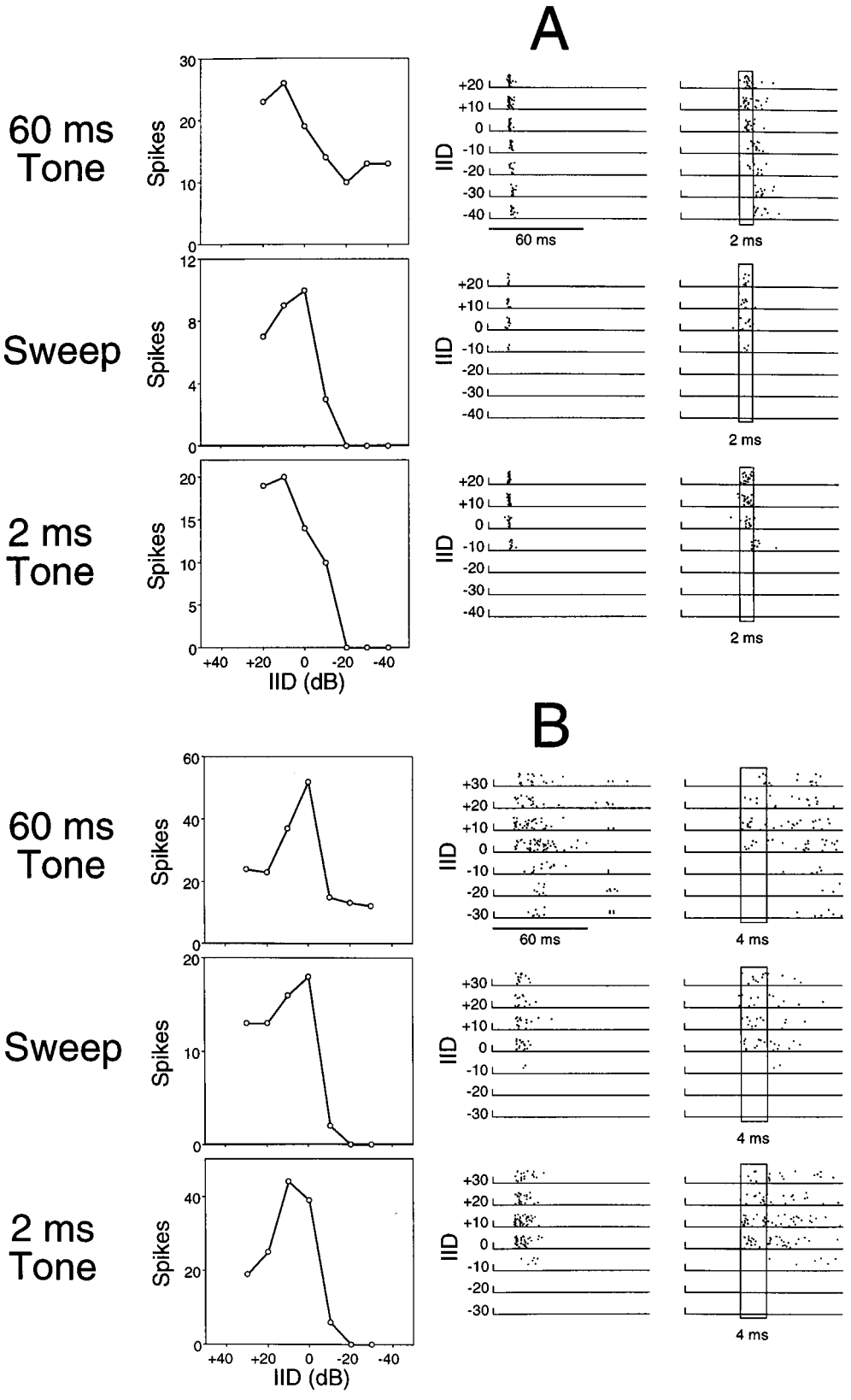

Figure 7. Two examples of Type B cells tested with 2 msec tones. The general format follows that of Figure 1. For both cells, the *top panel* shows responses to 60 msec tones, the *middle panel* shows responses to short sweeps, and the *bottom panel* shows responses to 2 msec tones. Note that the IID functions of the cells were partially inhibited for 60 msec tones and that they were completely inhibited for both of the short stimuli, the sweeps, and the 2 msec tones.

displayed by Type B cells changed with the duration of the stimulus.

We must point out that with the longer stimuli, Type B cells varied in how the spike counts of their later response components changed with IID. The later components of the majority of Type B cells appeared to be insensitive to IIDs (Fig. 5*A,B*), whereas the later components of some (14 of 54) Type B cells showed a substantial sensitivity to IIDs (Fig. 1*G*). Nevertheless, each of the

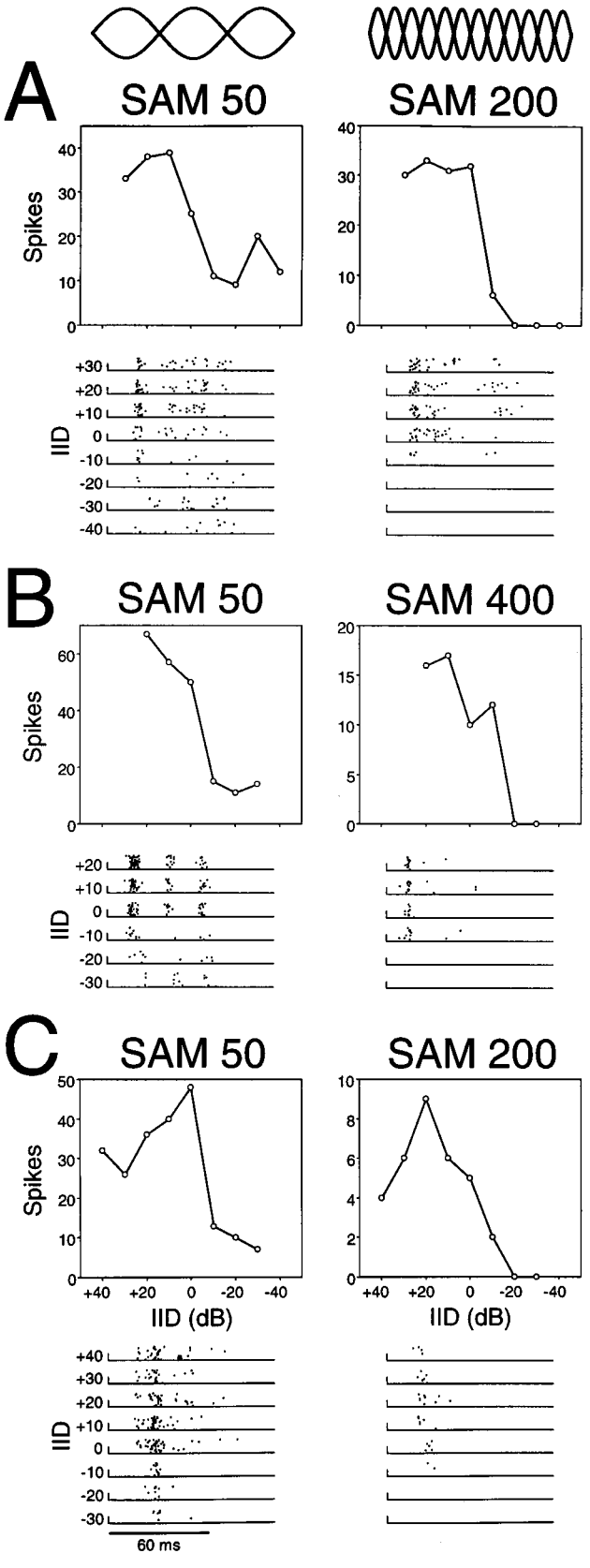

Figure 8. IID functions and raster plots from three Type B cells tested with two SAM rates. In all cases the carrier frequency was set to the cell's characteristic frequency. See Results for more detail.

Type B cells had later response components that displayed some degree of spike activity that was never completely inhibited at any IID tested.

Spike count versus response latency for longer stimuli: effects of the transient inhibition on response latency

The IID sensitivity of single neurons has traditionally been assessed by documenting how total spike counts change as a function of IID. Indeed, many IID-sensitive cells show a sharp sensitivity for IIDs based on total spike counts (Type A cells), and this response type is associated with some proportion of cells in a wide variety of auditory centers (Irvine, 1986; Kuwada and Yin, 1987; Sanes and Rubel, 1988; Park and Pollak, 1993; Irvine et al., 1996; Park et al., 1997). However, there are also many cells that, based on total spike counts, appear to have a much broader sensitivity to IIDs in that their IID functions are relatively shallow and do not decline to zero spikes (Irvine and Gago, 1990; Park, 1998; Wenstrup et al., 1988). Although the spike counts of these cells can still carry some important localization information, these cells have been viewed as being less suitable for coding a sound's location compared with Type A cells. However, in a recent report, Middlebrooks et al. (1994) showed that response parameters other than total spike count can carry important information about a sound's location. Middlebrooks et al. (1994) studied the temporal response patterns of cells in the auditory cortex using free-field stimulation that provided interaural timing cues as well as IIDs, whereas we used ear phones and focused only on IIDs. Nevertheless, the idea that response features other than total spike counts can carry localization information is relevant to the Type B cells we studied in that Type B cells may also code localization information in ways other than total spike counts. In the case of Type B cells, the transient inhibitory input silenced the initial spikes for IIDs favoring the inhibitory ear, and thus the response latency of those cells varied as a function of IID.

Transformations within the IC

An important question in auditory neuroscience has concerned the nature of hierarchical transformations in processing between the lateral superior olive (LSO) and the IC. The LSO is a prominent lower nucleus, dominated by IID-sensitive cells, that sends a heavy projection to the IC (Boudreau and Tsuchitani, 1968; Saint Marie and Baker, 1990; Glendenning et al., 1992; Park et al., 1996, 1997). Recent pharmacological and *in vivo* patch-clamp studies have intensified this question by showing that for many IC cells, IID sensitivity is shaped and in some cases created *de novo* within the IC, despite the projection from the LSO (Brownell et al., 1979; Park, 1988; Faingold et al., 1989, 1993; Li and Kelly, 1992; Vater et al., 1992; Park and Pollak, 1993; Klug et al., 1995; Covey et al., 1996; Kuwada et al., 1997). Why modify or recreate IID sensitivity in the IC when many lower cells are already highly sensitive to IIDs? The answer that appears to be emerging is that IID-sensitive cells in the IC resemble LSO cells only superficially. For example, two recent findings show how processing within the IC generates IID-sensitive cells with substantially different response properties from LSO cells. The first showed that many IC cells have a pronounced facilitation for particular IIDs, making them much more selective for IIDs compared with LSO cells (Semple and Kitzes, 1987, 1993; Irvine and Gago, 1990; Park and Pollak, 1993, 1994). The second finding showed that many IC cells have a persistent inhibition that substantially outlasts stimulus duration, affecting processing of sub-

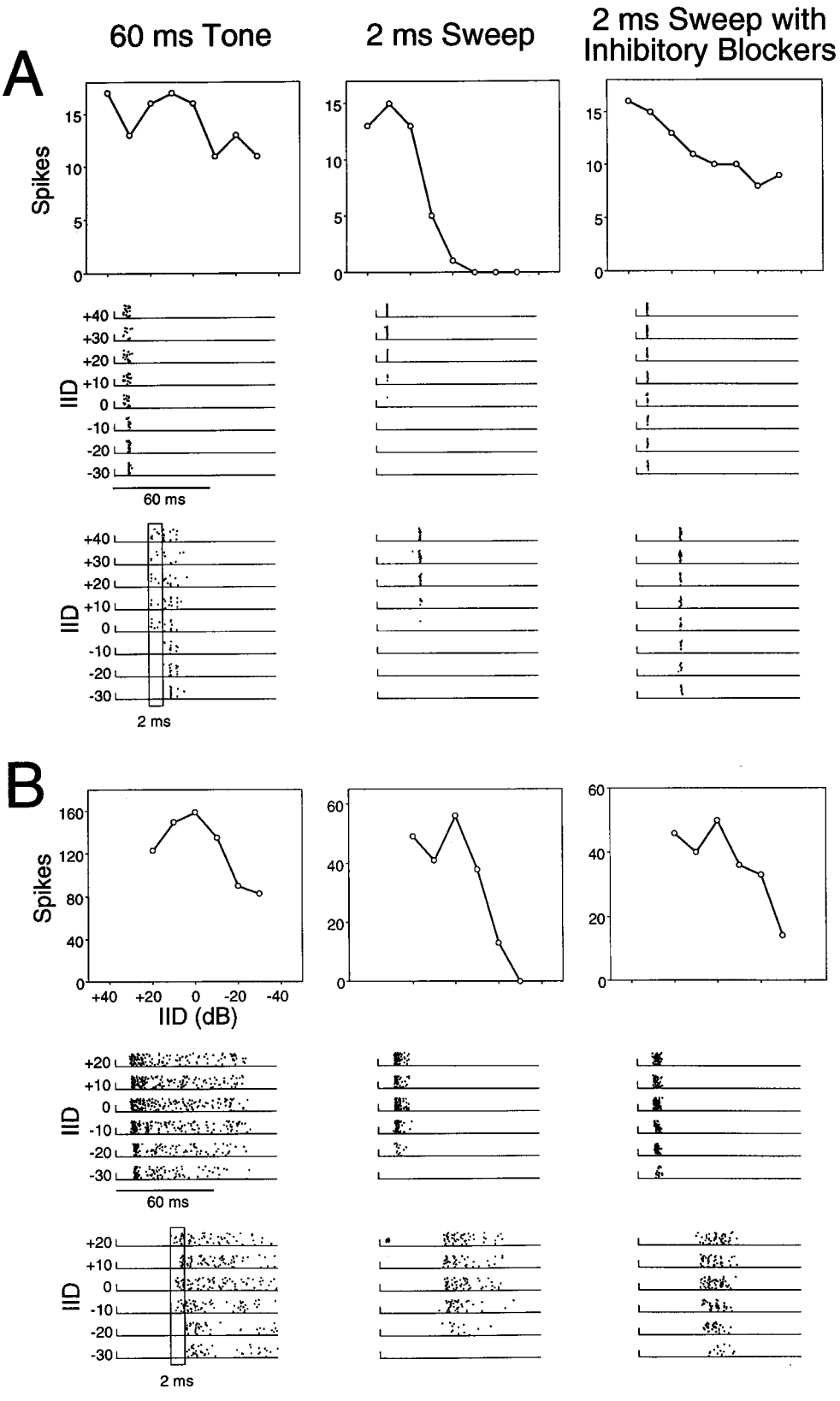

Figure 9. Two examples of Type B cells tested with 60 msec tones and with short sweeps before local application of inhibitory transmitter antagonists, and with sweeps during application of the antagonists. Note that the antagonists substantially blocked the early inhibitory component characteristic of Type B cells. See Results for more detail.

sequent stimuli (Carney and Yin, 1989; Yin, 1994; Kidd and Kelly, 1996; Klug et al., 1997). The present study identifies yet another major difference between LSO and IC cells in that many IC cells have Type B response patterns.

The differences described above between LSO and IC cells are consistent with their innervation patterns. The input pattern to LSO cells is relatively simple: they receive prominent excitatory inputs from the ipsilateral cochlear nucleus and prominent inhibitory inputs from the contralateral cochlear nucleus via a connection through an intermediate, inhibitory nucleus, the medial nu-

cleus of the trapezoid body (Cant and Casseday, 1986; Sanes and Rubel, 1988; Glendenning et al., 1992). In contrast, IC cells integrate many more excitatory and inhibitory inputs from numerous lower centers (Pollak et al., 1986; Winer et al., 1995; Oliver et al., 1997), providing opportunities for more complex output patterns.

Future directions

Future studies will examine two main issues. The first is whether Type B cells are specific to bats or whether they are common among mammals in general. Although cells with shallow IID functions that do not decline to zero spikes have been reported in cats, the underlying circuitry that shapes the partially inhibited response pattern of those cells has not yet been reported. The second issue we plan to examine concerns how Type B cells might function in the auditory world of the free-tailed bat. One aspect of the bat's auditory world concerns echolocation. Free-tailed bats emit sweep-like vocalizations and localize the echoes of those calls that return from flying insects. While searching for prey, they use long, shallowly sweeping calls. As they close in, they use progressively shorter, more rapidly sweeping calls (resembling the sweeps in our study). It is thought that the long calls are better-suited for detecting and identifying prey, whereas the short calls are better for accurate, rapid-fire localization (Young, 1989). Our present results suggest that Type B cells might respond in a manner consistent with these different signals and their related behavioral tasks. It may be that the long calls, associated with target detection, evoke partially inhibited IID functions, providing at least some response regardless of target location. On the other hand, the short calls, associated with localization of targets during the terminal, interception phase of hunting, evoke completely inhibited IID functions that may be better-suited for fine grain localization. Another aspect of the bat's auditory world concerns intraspecific communication calls, which include long (~60 msec) tone-like calls, as well as complex amplitude and frequency-modulated calls (Park et al., 1998). Again, the way in which Type B cells respond to the location of these calls may depend on call characteristics. Testing Type B cells with digitized echolocation and communication calls will help elucidate the functional role of these cells in localizing the various signals that the bats hear.

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