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# Transient Frequency and Intensity Sensitivities of Central Auditory Neurons Determined with Sweep Tone

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## Abstract

**To determine the transient frequency and intensity sensitivities of central auditory neurons, we implemented an exponential sweep tone stimulus (2 sec in period, mean sweep rate 3.3 octave/sec), intensity of which varied systematically across trials. Response of single units to the stimulus was studied at the inferior colliculus (IC) of urethane-anesthetized rats. Most IC units responded to the sweep tone by one or more transient increases in discharge rate. The area of increased discharge, or response area (RA), was delineated on the frequency-intensity plane. The tip of RA gives the best frequency (BF) and minimum threshold (MT) of the cell. We also compared the BF and MT concurrently obtained with another method, viz., the conventional 'audio-visual' method of subjective judgment. Results showed that for the same population of cells (n=130), correlation between the two methods is better for BF (r=0.91) than for MT (r=0.78). Such discrepancy was discussed in relation to the response characteristics of these central auditory neurons.**

**Key Words: best frequency, minimum threshold, response area, inferior colliculus, rat**

## Introduction

In the auditory periphery, single auditory nerve fibers all responded to pure tone stimuli (9). Their frequency tuning is characterized by the so-called 'characteristic frequency' (CF) and 'minimum threshold' (MT), marking the tip of the tuning curve (6). Tuning curve is conventionally generated with tone bursts changing systematically in discrete frequency and intensity steps (13). In the central auditory system like the primary cortex (4), MT and CF (or more often called best frequency, BF; 21, 23) are determined basically in the same manner like the auditory periphery. A number of studies on central auditory neurons (18, 22) showed that steady tones are actually less effective stimuli for these cells. Many midbrain neurons, for example, will not respond to a steady tone but will rather to a time-varying stimulus. Specifically, these cells tend to respond at the onset of an amplitude modulated (AM) signal (1, 17), or a frequency modulated (FM) tone (14, 15, 17, 19). In view of such dynamic response properties, an alternative method to characterize such auditory sensitivities is in demand. This is particularly important in the study of speech sound coding in the central auditory system as higher order neurons are often driven by complex sounds rather than by pure tones.

In determining BF and MT of central auditory neurons, the traditional stimulus with tone burst is commonly used. This stimulus has problems in at least three aspects: (a) discrete frequency steps produce an inaccurate BF estimate, particularly when cells display extremely fine frequency tuning (10), (b) a tone burst has a rise-fall time, and it will inevitably introduce an AM component, that is known to strongly affect the response of central auditory neurons (16), and (c) in case the cell does not respond to steady tones (14, 15, 20, 22) their sensitivity will not be determined.

In this study, we implemented a sweep tone that varies systematically in both frequency and intensity, so that the neuron's response area (RA) can be scanned efficiently. This tone sweep is fast enough to activate most cells including those not driven by steady tones,

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but slow enough not to contaminate with the FM response of the cells. We further developed a method of constructing the cell's RA and subsequently deriving its BF and MT. For comparison, we determined BF and MT for the same population of neurons according to the conventional approach–the 'audio-visual' method (i.e., subjective judgment of the best frequency and lowest intensity of a tone that can evoke a unit's minimal response). Results showed that our tone sweep is a simple but efficient method of estimating BF and MT of many central auditory neurons.

## Materials and Methods

A total of 30 Sprague-Dawley rats (body wt 150-250 gm) were used. The surgical procedure has been described in pervious studies (14). In brief, animals were anesthetized with urethane (2.0 gm/kg, i.p.). The skull was exposed on the dorsal side and a 5-mm fenestration drilled on the skull overlying the occipital lobe. Dura was resected and the cortex overlying the IC exposed. The rat was then transferred to a sound-treated room (IAC) and the head fixed to a special holder by means of a screw cemented to the frontal skull. Acoustic stimulation was delivered from a free field speaker (Pioneer SP77) placed 70 cm along the acoustic axis contralateral to the side of recording (30° in azimuth, 0° in elevation). A micropipette electrode (30 MegOhm) was remotely controlled by a microdrive (Narishige) and advanced through the occipital lobe to hunt for single auditory units in the midbrain. Units were identified by either their spontaneous activities or more often their time-locked response to a hunting click (100 μsec rectangular pulse) with short latencies (10-20 msec). After unit isolation, a computer-synthesized waveform was generated (Tucker Davis Technology DD1) at a time resolution of 0.4 msec and 16 bit in vertical resolution. The waveform consisted of an exponential rise-and-fall over a 2-sec period (see Fig. 1). The signal is a modulator that controlled the instantaneous frequency of a function generator (Tektronix FG 280) by its voltage-control-frequency input. The output of the function generator, now in the form of a slow tone sweep, was then used as the input to the speaker. The frequency response of the acoustic delivery system is reasonably flat (±12 dB from 0.4-40 kHz). Unit responses were recorded by sampling the extra-cellular action potential preconditioned to a rectangular pulse (0.4 msec wide).

Recording sites in the brain were identified histologically at the end of experiment by tracing the electrode track in the IC after sacrificing the rat with an over dose of urethane. A successful recording was always found to penetrate the IC, through its central or external nucleus.

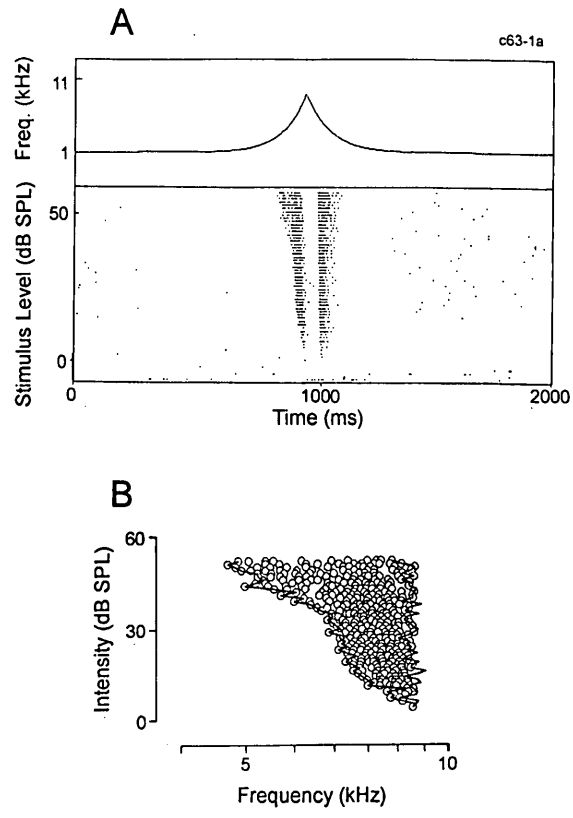


Fig. 1. (A) The single band response area (RA) of an IC neuron obtained with a slow up-down sweep tone. Note the tone sweeps across the cell's RA twice in each stimulus presentation (upper panel), resulting in two rather symmetrical RAs (lower panel). The stimulus level attenuates systematically over 60 trials and spike responses of the cell displayed in dot raster. Isolated dots in lower panel are likely spontaneous activities unrelated to the stimulus. (B) The RA to the up-sweep shown in greater details with its boundary (solid line) extracted by our analysis program. Each circle represents a single spike. The tip of RA marks the cell's minimum threshold, MT (y-coordinate) and best frequency, BF (x-coordinate).

The RA of the cell was displayed on the frequency-intensity plane together with the occurrence of all spikes. Fig. 1 shows a representative response of an IC cell. A software program determined the most sensitive frequency (BF) and the minimal intensity (MT) of the tone evoking spikes. The program basically extracts the boundary of the activity pack (RA) in the frequency-intensity plane by systematically moving a rectangular window over the dot-raster using a threshold value set above the cell's spontaneous activity level. The co-ordinates of the RA tip give the BF and MT of the cell.

For audio-visual determination of BF and MT, the same speaker was used to deliver a continuous tone. Minimal response was determined subjectively by manually adjusting the tone frequency (by turning the digital frequency dial of the function generator)

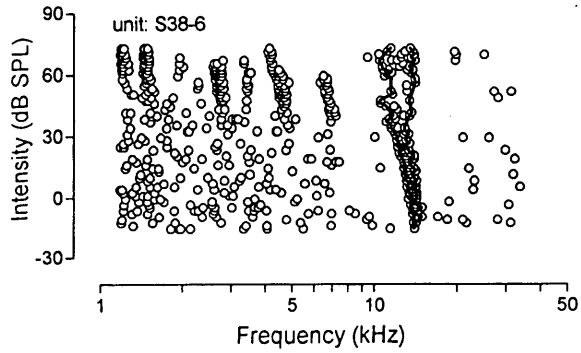


Fig. 2. Multiple-band RA of an IC neuron. Caption similar to Fig. 1B. Dots are replaced by circles to represent spike discharges. Isolated circles are spontaneous discharges ignored by the analysis program. Note the boundary of RA was only extracted for the most sensitive peak.

and attenuating its intensity (with a decade attenuator, HP 350D) in step of 1 dB until the cell no longer responded.

## Results

Of a total of 150 units recorded, 135 (90%) responded to the hunting click and 15 (10%) were spontaneously active. The majority of them (93%, n=140) responded to the 2-sec exponential tone sweep. BFs found along the course of electrode penetration, especially through the central IC, were quite consistent with the low-to-high distribution well-known for this animal. Of the 130 units collected with complete data sets (which form the results we shall present), about one-third (37%) displayed a single band RA similar to that shown in Fig. 1, while the rest (63%) displayed more complicated features like multiple bands (Fig. 2). Regardless of the complexity of RA, there was always a single value of BF-MT extractable by our analysis program.

The effects of varying the speed of tone sweep were tested in a small sample of units (n=20). Choosing a modulation waveform of duration 1, 4 or 8 sec instead of 2 produced no significant change in the RA. With a longer period, cells usually responded with more spikes and details in the RA better revealed. Some cells however failed to respond to faster sweeps. Preliminary data suggested that the 2 sec likely represents an optimal period in evoking spikes from the population of IC cells studied. We further tested the effects of different modulation waveforms, viz., an exponential versus a linear ramp. In general, the exponential envelop delineated finer details of the RA, likely related to the logarithmic arrangement of frequencies at the basilar membrane (13). For these reasons, the exponential sweep tone of 2-sec period was chosen as the standard stimulus in this experiment.

Selecting a carrier frequency slightly off the BF of the cell usually did not change the values of BF and MT so calculated. As a standard procedure, a frequency range centered roughly at the cell's BF (as determined audio-visually) was chosen together with sweep range as large as possible as allowed by the function generator. The range actually used ranged 1.9 to 7.0 in octave/sec (mean = 3.3 octave/sec).

For efficient data collection and reproducibility, the number of stimulus repetitions was set to 60, during which stimulus level was varied systematically. To cover the dynamic range of the cell, the attenuation step used ranged 0.2 to 1.5 dB (mean=0.9 dB), which was quite sufficient to cover most cells' dynamic range of response of about 30 dB. BF and MT were minimally affected by the size of attenuation step. Intensity steps across trials chosen as an increase of stimulus level instead of attenuation also had little effects on MT and BF, likely related to the rather long 2-sec cycle.

One important determinant of the response is the direction (i.e., increase or decrease in frequency) of tone sweep. All cells responded to the up-sweep (i.e., an increase in frequency), but only 90% (n=117) to the down-sweep. For the 117 cells responded to both directional sweeps, most of them (95%) showed asymmetrical response patterns. The slight preference of response to the up-sweep could be related to adaptation of the cells to successive stimuli. Since the up-sweep always precedes the down-sweep. Changing the sweep sequence from up-down to down-up could change the symmetry of the response. For practical purposes, calculation of BF and MT was based exclusively on the cell's response to the up-sweep. In BF estimation, the central transmission delay of individual cells (22) was taken into account.

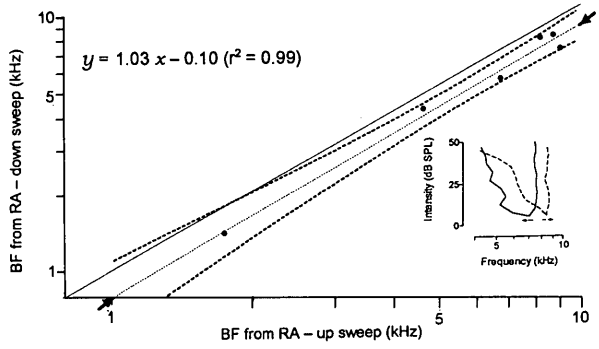


Fig. 3. Comparison of BFs (filled circles) based on either up-sweep or down-sweep data from 6 units. The thin dotted line (arrows) shows the linear regression (equation) and 95% confidence interval (dashed lines). For comparison, the diagonal (solid line) is also plotted. The inset shows the two RAs from one of the 6 units. The small arrows indicate the direction of the tone sweep. Note the up-sweep displays a BF of higher frequency.

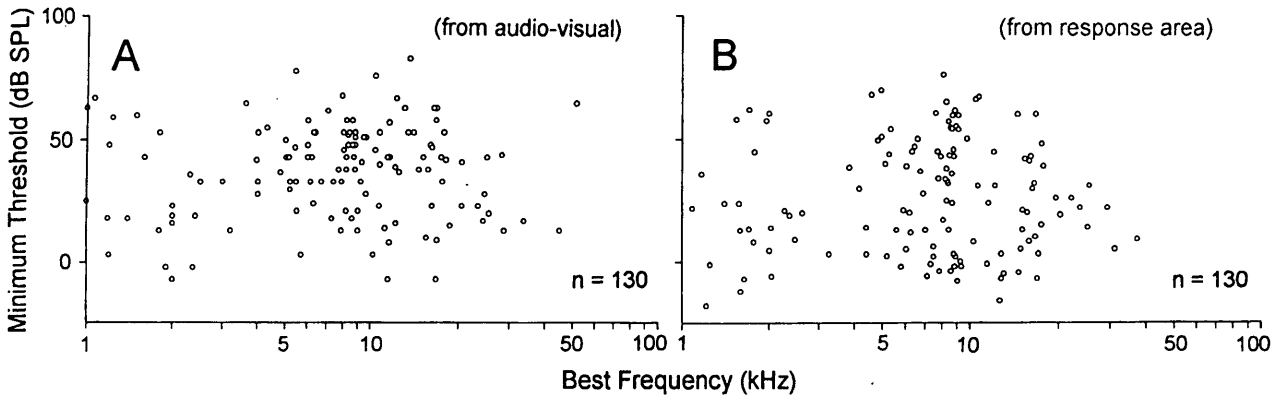


Fig. 4. Scatter plot of BF and MT determined by the audio-visual method (A) and the RA method (B). Note some circles overlap. Same population of cells in A and B.

We assumed that whenever the cell discharged a driven spike, it was responding to that part of the stimulus beforehand by a time lag equal to the central transmission delay. This was simply accounted for by subtracting the click response latency of the cell. For about 10% of the cells that did not respond to the hunting click, a value equal to the population mean (13 msec) was used.

For a small number of cells (n=6) that responded symmetrically to the up- and down-sweep, we estimated the discrepancy in BF estimation based on up-sweep or down-sweep data. From the limited sample, up-sweep data all gave a slightly higher estimate in BF (Fig. 3). We explain this by a longer response latency at threshold stimulus levels. Since at MT intensity, the stimulus to the cell will be delivered at threshold level, resulted in a slightly longer transmission delay. For the up-sweep data, the tip of RA will be displaced to the right-hand-side of the RA, giving rise to a BF estimate higher in frequency than the actual one. Whereas for the down-sweep data, the same displacement towards the right-hand-side will result in a BF lower in frequency.

For comparative viewing of the population data, BFs and MTs of the 130 IC units determined by the two different methods are displayed side-by-side in a scatter plot (Fig. 4) and in histograms (Fig. 5). For the RA data, BFs were found between 1.1 to 56.2 kHz, with a peak at 8.9 kHz (Fig. 5B). These results are consistent with both the rat's electro-physiological data (14) and its behavior (2). The mean departure in BF (0.15 octave, SD=0.12 octave) without taking into account the transmission delay is not insignificant. For instance, 1.0 kHz will have been corrected to 0.903 kHz with delay compensation. However, compensating transmission delay or not had little effects on the overall shape of the histograms. This observation can be explained by the relatively large bin size used in generating the histogram.

In comparison, the BFs estimated with audio-visual method covered a slightly narrower range (1.1 to 35.5 kHz), with fewer cells at the high frequency end. The peak was again centered at 8.9 kHz (Fig. 5A). The two distributions (Fig. 5 A, B) are not statistically different ($p$=0.6, Student's $t$-test).

The relationship of BF derived from RA versus audio-visual method is displayed on a regression plot (Fig. 6). The fact that the regression line nearly overlaps with the diagonal and the intercept is almost zero indicating a linear relation between the two data sets.

The MTs derived from the RA data (Fig. 5D) spread from -5 to 45 dB SPL with a flat plateau. The distribution of MT is almost equal probability from -15 to 75 dB SPL. In comparison, the audio-visual MTs (Fig. 5C) occupy a smaller intensity range (-5 to 75 dB SPL) and are less uniform in distribution with a peak at a higher intensity (45 dB SPL). Contrary to BF, the two MT distributions are statistically different ($p$<0.01, Student's $t$-test). The regression plot of the two MT data sets (Fig. 7) confirmed the rather poor correlation between the two data sets, apparently related to an over-estimate of MT by the audio-visual method.

In spite of the above discrepancies, the distributions of audio-visual BFs and MTs we found in this experiment are consistent with the previous reports from our laboratory (14).

## Discussion

Even though nearly 25% of IC cells responded to click are reported not driven by steady tones (15), a principal finding of this study is almost all IC neurons (93%) have responded to our 2-sec exponential sweep tone. This indicates that the tone sweep is indeed an effective stimulus for determining their transient frequency and intensity sensitivities of

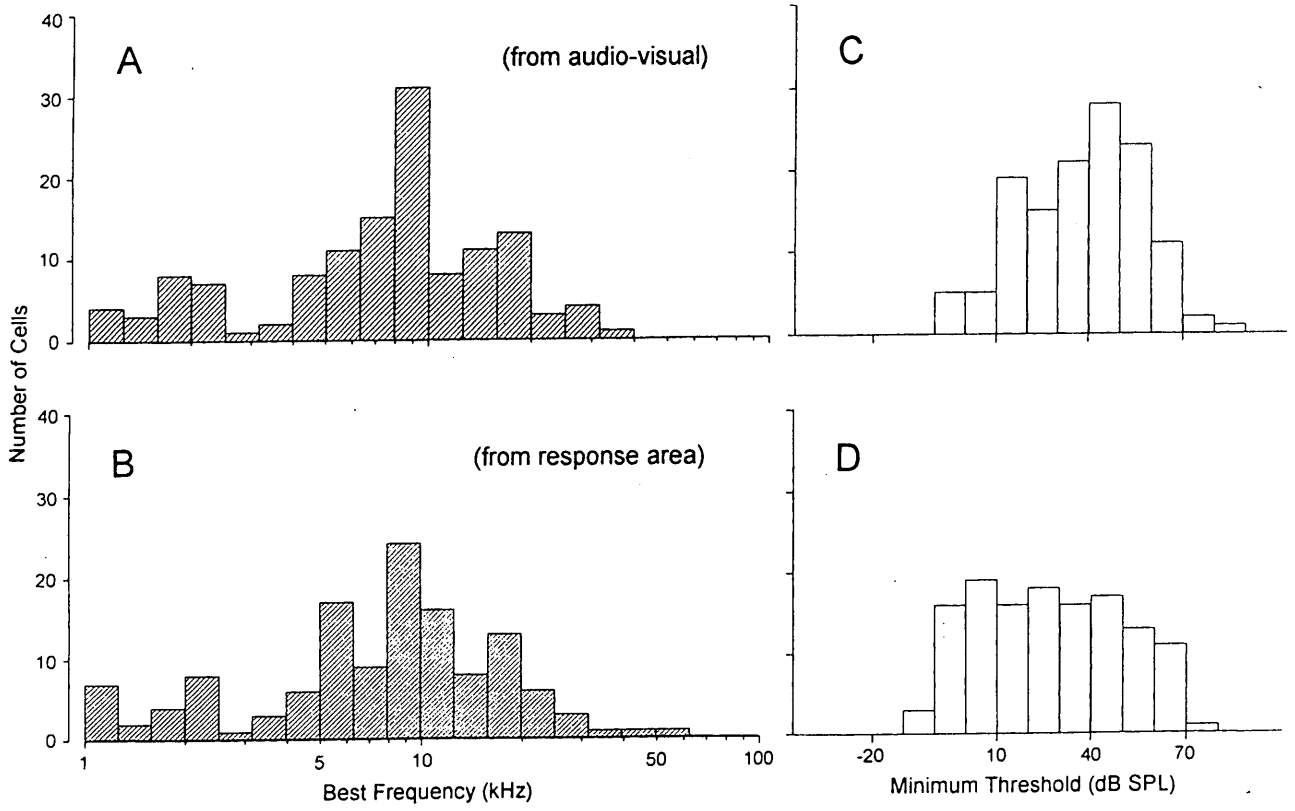


Fig. 5. The histograms of BF and MT by the audio-visual method (A, C) and by RA method (B, D). Same data as in Fig 4.

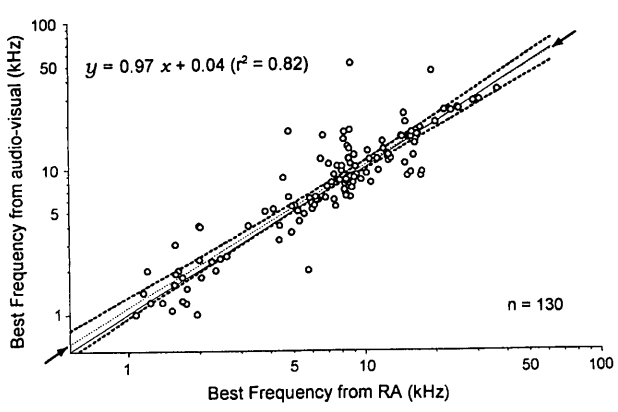


Fig. 6. Regression plot of BFs derived from the method of RA or audio-visual. Thin dotted line (arrows) is the linear regression (equation) and the dashed lines, 95% confidence interval. Note the overlap with the diagonal (solid line) indicating a good correlation between the two data sets.

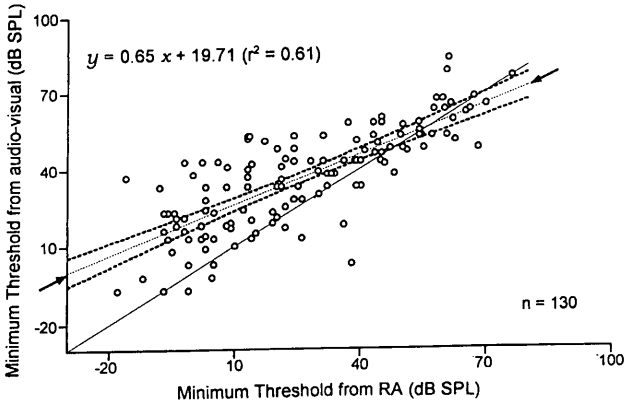


Fig. 7. Regression plot of MTs derived from the method of RA or audio-visual. Caption similar to Fig. 6. Note the departure from the diagonal (solid line) indicating over-estimate of MT with the audio-visual method.

the midbrain neurons. The fact that FM sensitivities are prevailing in the IC did not handicap much of the usefulness of this approach. Since the major FM sensitivity at the IC was found between 10 to 20 Hz (7, 14, 15, 17), equivalent to a 0.05 to 0.1 sec in period, which is much higher in modulation rate than the 2-sec sweep tone we used.

This study also the first systemic attempt to compare a quantitative method versus an audio-visual one. While BFs are comparable, the audio-visual method on the average produced over-estimated MTs. The discrepancy can be readily explained by adaptation of neurons to the continuous tone, as it was the case with the audio-visual method.

We did not attempt to compare the sensitivities obtained by audio-visual and those obtained by short tone bursts. Since a systematic evaluation of the

effects of rise-fall time of the tone envelop will be too time-consuming and that comparison should belong to a separate study.

Since our present method of frequency scanning is a continuous one, important advantages of this stimulation are its capability (a) to reveal more accurately the cell's BF, (b) to reveal the complexity of the RA which may well be concealed by discrete frequency and intensity steps. The fact that the details of RA were satisfactorily revealed (see Fig. 2) suggested that the discrete intensity steps we used (n=60 over an intensity range of about 30 dB) are sufficient for most IC cells. However, we did not attempt to elaborate on the complexity of RA of IC neurons as that is beyond the scope of this study. Furthermore, the relatively short data collection time (2 minutes) in characterizing a cell's BF and MT appeared to be practical as most IC units can be maintained in a stable recording situation for about 10 minutes.

The fact that IC neurons and cortical neurons may not respond to pure tones poses an important barrier for effective determination of transient frequency and intensity sensitivities of these cells. In the central auditory literature, there is still an overwhelming tendency to adopt the approach used in studying the auditory periphery. For instance, the tuning curve approach is widely used in characterizing transient sensitivities even at the cortical level (4). We know that pure tone evokes only a transient response of auditory cortical neurons at the tone onset and often not during the steady state of the stimulus. It is now clear that AM sensitivity is also rather prevailing in the central auditory system staring from the cochlear nucleus to the midbrain (3, 5, 8, 11, 24). The need of an alternative method to assess central neurons' sensitivity is in order. This sweep tone approach provides an alternative solution to this problem.

## Acknowledgments

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