GABAergic Disinhibition Affects Responses of Bat Inferior Collicular Neurons to Temporally Patterned Sound Pulses

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Lu, Yong, Philip H.-S. Jen, and Min Wu. GABAergic disinhibition affects responses of bat inferior collicular neurons to temporally patterned sound pulses. J. Neurophysiol. 79: 2303–2315, 1998. Using the big brown bat, Eptesicus fuscus, as a model mammalian auditory system, we studied the effect of GABAergic disinhibition by bicuculline on the responses of inferior collicular (IC) neurons to temporally patterned trains of sound pulses delivered at different pulse repetition rates (PRRs) under free-field stimulation conditions. All 66 neurons isolated from eight bats either discharged one to two impulses (phasic on responders, \( n = 41, 62\% \)), three to eight impulses (phasic bursters, \( n = 19, 29\% \)), or many and long impulses throughout the entire duration of the stimulus (tonic responders, \( n = 6, 9\% \)). Whereas 50 neurons responded vigorously to frequency-modulated (FM) pulses, 16 responded poorly or not at all to FM pulses. Bicuculline application increased the number of impulses of all 66 neurons in response to 4 ms pulses by 15–1425%. The application also changed most phasic on responders into phasic bursters or tonic responders, resulting in 12 (18%) phasic on responders, 34 (52%) phasic bursters, and 20 (30%) tonic responders. Response latencies of these neurons were either shortened (mean = 0.5–6.0 ms, lengthened (mean = 9, 14%) by 0.5–2.5 ms or not changed (mean = 32, 48%) on bicuculline application. Each neuron had a highest response repetition rate beyond which the neuron failed to respond. Bicuculline application increased the highest response repetition rates of 62 (94%) neurons. According to total discharge rate-based modulation transfer functions (total rate MTFs), 66 neurons had low-pass filtering characteristics both before and after bicuculline application. According to total discharge rate-based modulation transfer functions (total rate MTFs), filtering characteristics of these neurons can be described as band-pass (mean = 52, 79%), low-pass (mean = 12, 18%), or high-pass (mean = 2, 3%) before bicuculline application. Bicuculline application changed the filtering characteristics of 14 (21%) neurons. According to synchronization coefficient-based modulation transfer functions, filtering characteristics of these neurons can be described as low-pass (mean = 41, 62%), all-pass (mean = 11, 17%), band-suppression (mean = 7, 10.5%), and band-suppression–band-pass filters (mean = 7, 10.5%). Bicuculline application changed filtering characteristics of 19 (29%) neurons.

INTRODUCTION

The importance of temporally patterned sound pulses in acoustic communication and/or orientation of animals has been demonstrated in previous behavioral and neurophysiological studies (Busnel 1976). For example, frogs use spectral cues for communication and temporal cues [pulse shape, pulse repetition rate (PRR), duration, and rise-decay times] for call discrimination (Capranica 1966). Similarly, echolocating bats vary pulse duration, pulse intensity, and PRR during different phases of hunting to ensure successful prey capture (Griffin 1958; Simmons et al. 1979). Previous studies have shown that temporally patterned trains of sound pulses delivered at different PRRs affect the sensitivity of auditory neurons. Whereas individual auditory neurons show different filtering properties to PRRs (Condon et al. 1994; Jen and Schlegel 1982; Pinheiro et al. 1991; Wu and Jen 1995; Wu et al. 1996), increasing PRR sharpens the auditory directional sensitivity (Wu and Jen 1996) and increases the minimum threshold (MT) and response latency of auditory neurons (Chen and Jen 1994; Donaldson and Rubel 1990; Phillips et al. 1989). In the ascending auditory pathway, the inferior colliculus (IC) receives excitatory and inhibitory inputs from all lower auditory nuclei (Adams 1979; Adams and Mugnaini 1984; Casseday and Covey 1992, 1995, 1996; Covey and Casseday 1986; Massopust and Ordy 1962; Oliver et al. 1994; Pollak and Casseday 1989; Roberts and Ribak 1987a,b; Rockel and Jones 1973; Saint Marie et al. 1989; Shneiderman and Oliver 1989; Zook and Casseday 1987; Zook et al. 1985). The IC also receives descending inputs from the auditory cortex through the cortico-collicular pathways (Hefti et al. 1995; Huffman and Henson 1990; Syka et al. 1988). These excitatory and inhibitory inputs contribute importantly to auditory temporal processing in this nucleus (Casseday and Covey 1995). Neurotransmitters that mediate the inhibitory inputs to IC neurons are either \( \gamma \)-aminobutyric acid (GABA) or glycine (Casseday et al. 1994; Fubara et al. 1996; Johnson 1993; Park and Pollak 1993a,b; Vater et al. 1992).

Previous studies have shown that iontophoretic application of GABA inhibited spontaneous or noise-evoked activity and decreased the number of impulses of IC neurons in response to sound stimuli. In contrast, application of bicuculline, which is an antagonist for GABA\( _{A} \) receptors (Borman 1988; Cooper et al. 1982), increased the number of impulses, changed discharge patterns, and shortened the response latency of IC neurons (Ebert and Ostwalt 1995; Faingold et al. 1989, 1991; Palombi and Caspary 1992; Park and Pollak 1993a,b; Vater et al. 1992; Yang et al. 1992). We recently demonstrated that GABAergic disinhibition by bicuculline application shortened the recovery cycles of IC neurons (Lu et al. 1997). Because the recovery cycle is an important neuronal property that determines a neuron’s ability to respond to sound pulses delivered at different interpulse intervals or PRRs, we hypothesize that shortening a neuron’s


TABLE 1. Relationship among the pulse repetition rate (PRR), the pulse duration (PD), the interpulse interval (IPI), and the pulse number (PN) in the 300-ms pulse trains

<table>
<thead>
<tr>
<th>PRR, pps</th>
<th>PD, ms</th>
<th>IPI, ms</th>
<th>PN</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>4</td>
<td>1,000</td>
<td>1</td>
</tr>
<tr>
<td>4</td>
<td>4</td>
<td>250</td>
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</tr>
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<td>67</td>
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<td>4</td>
<td>12</td>
<td>24</td>
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<tr>
<td>100</td>
<td>4</td>
<td>10</td>
<td>29</td>
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<tr>
<td>120</td>
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<td>8</td>
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<tr>
<td>150</td>
<td>4</td>
<td>7</td>
<td>44</td>
</tr>
<tr>
<td>200</td>
<td>4</td>
<td>5</td>
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<tr>
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<td>4</td>
<td>74</td>
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<tr>
<td>400</td>
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<td>119</td>
</tr>
<tr>
<td>500</td>
<td>2</td>
<td>2</td>
<td>149</td>
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</table>

We report here that most IC neurons studied had band-pass total rate MTF filtering properties but had low-pass average rate MTF and sync MTF filtering characteristics to PRR. Bicuculline application increased the number of impulses of all IC neurons in response to each presented pulse at each PRR. Each neuron had a highest response repetition rate beyond which the neuron failed to respond. Bicuculline application increased the highest response repetition rates of 62 (94%) neurons studied. However, bicuculline application did not change the filtering characteristics of total rate MTFs, average rate MTFs and sync MTFs of most IC neurons.

METHODS

Eight big brown bats, *Eptesicus fuscus*, (5 males, 3 females, 15–24 g, body wt) were used for this study. The surgical procedures were basically the same as in previous studies (Jen et al. 1987, 1989). Briefly, under pentobarbital sodium (Nembutal) anesthesia (45–50 mg/kg body wt), the flat head of a 1.8 cm nail was glued onto the exposed skull of each bat with acrylic glue and dental cement 1 or 2 days before the recording session. Exposed tissue was treated with an antibiotic (Neosporin) to prevent inflammation. During recording, the bat was administered the non-steroidal anti-inflammatory drug, Innovar-Vet (Fentanyl 0.08 mg/kg body wt, Droperidol 4 mg/kg body wt), and put inside a bat holder (made of wire mesh) that was suspended in an elastic sling. The bat’s head was immobilized by fixing the shank of the nail into a metal rod with a set screw (Suga and Schlegel 1972). The head was oriented with the eye-snout line pointed to 0° in azimuth and 0° in elevation of the bat’s frontal auditory space. Small holes were made in the skull above the IC for insertion of electrodes to record sound activated neural responses. Recordings were conducted inside a double-wall sound proof room (temperature 28–30°C) with its ceiling and inside walls covered with 3-in convoluted polyurethan foam to reduce echoes. Recording depths of IC neurons were read from the scale of a microdrive (David Kopf model 640). [The experiments were conducted in compliance with National Institutes of Health publication 85-23, *Principles of Laboratory Animal Care*.

![Figure 1A](http://jn.physiology.org/)

**FIG. 1.** A: comparison of response latency of 66 inferior collicular (IC) neurons before (○, lat) and after (●, lat bic) bicuculline application. Bicuculline application affected response latency of 34 neurons. ---, response latencies of collicular neurons that were ≤10 ms (n = 40, to the left of ---) or >10 ms (n = 26, to the right of ---). B: variation in response latency of 66 collicular neurons on bicuculline application. □, response latency was increased (positive) or decreased (negative) on bicuculline application; ■, no change in response latency on bicuculline application. Among 34 neurons the response latencies of which were affected by bicuculline application, all but 1 neuron with >10 ms response latency decreased the response latency on bicuculline application. In contrast, neurons with response latencies <10 ms either increased or decreased response latency on bicuculline application.
and with the approval of the Institutional Animal Care and Use Committee (1438) of the University of Missouri-Columbia.

The electronic instruments used to generate acoustic stimuli were the same as those used in recent studies (Jen et al. 1987, 1989; Kamada et al. 1992). Briefly, continuous sine waves from an oscillator (KH model 1200) were formed into 4-ms tone pulses (0.5 ms rise-decay times) by a homemade tone burst generator (electronic switch) driven by a stimulator (Grass S88). The tone pulses then were amplified after passing through a decade attenuator (HP 350D) before they were fed to a small condenser loudspeaker (AKG model CK 50, 1.5-cm diam, 1.2 g). The loudspeaker was placed 23 cm away from the bat and 30° contralateral to the recording site. The loudspeaker was calibrated with a Bruel and Kjær ½-in microphone (4135) placed at the bat’s ear. The output was expressed in dB SPL referred to 20 μPa root mean square.

A function characteristics curve was plotted for the loudspeaker to determine the maximal available stimulus intensity at each frequency.

When an IC neuron was isolated by 4-ms pulses, its best frequency (BF) and MT were determined by systematically changing the frequency and intensity of the sound pulses. At the MT, the neuron, on average, responded with 50% probability to BF pulses. As E. fuscus uses downward sweeping frequency modulated (FM) pulses for echolocation (Simmons et al. 1979), the neuron’s MT was redetermined with FM pulses sweeping one octave downward across the neuron’s BF. FM pulses then were used for subsequent studies. However, when a neuron did not respond or responded very poorly to FM pulses, BF pulses were used. Pulse intensity was set at 20–30 dB above a neuron’s MT.

A neuron’s responses to pulses delivered at different PRRs were studied with 300-ms pulse trains, which consisted of 4-ms FM or BF pulses. By delivering various numbers of pulses (1, 2, 3, 8, 12, 19, 24, 29, 35, 44, 59, or 74) at different interpulse intervals (1,000, 250, 167, 100, 40, 25, 15, 12, 10, 8, 7, 5, or 4 ms) within each pulse train, the neuron’s number of impulses in response to PRRs of 1, 4, 6, 10, 25, 40, 67, 83, 100, 120, 150, 200, or 250 pps was recorded sequentially. These pulse trains are pulsatile amplitude modulated trains (PAM) in which the amplitude of individual pulses was modulated at 100%. The pulse trains were delivered at two trains per second with a 200-ms silent period between trains. When a neuron’s responses to pulses delivered at PRRs higher than 250 pps were studied, pulse duration was reduced to 2 ms (0.25 ms rise-decay times). Table 1 shows the relationship among the PRR, pulse duration, interpulse interval, and pulse number within the 300-ms pulse trains.

The piggy-back multibarrel electrodes used to record acoustically evoked single neuron responses and to inject drugs iontophoretically to the recording site were the same as in our recent study (Lu et al. 1997). Each multibarrel electrode was composed of a three-barrel electrode (tip: 10–15 μm) “piggybacked” to a 3 M KCl single-barrel electrode (tip: <1 μm; impedance: 5–10 MΩ) in which the tip of which was extended ~10 μm from the tip of the three-barrel electrode. The 3 M KCl single-barrel recording electrode was connected by a silver wire to an amplifier (HP 465A) followed by an electronic filter (KH 3500). One of the barrels of a triple barrel electrode was filled with bicuculline methiodide (10 mM NaCl, pH 3.0, Sigma). The bicuculline was prepared just before each experiment, and the electrode filled immediately before use. The bicuculline channel was connected via silver-silver chloride wire to a microiontophoresis constant current generator (Medi- cal Systems Neurophore BH-2), which was used to generate and monitor iontophoretic currents. During bicuculline application, 1-s pulses of 40 nA at 0.5 pps were applied for 1 min before data acquisition. Application current then was changed to 10 nA during data acquisition. The other two barrels were filled with 1 M NaCl (pH 7.4), one of which was used as the ground and the other as the balanced barrel. The balance barrel was connected to balance module. The retaining current was negative 8–10 nA.

To determine any potential artifacts due to passing current or low pH values, the balanced barrel was filled with 1 M NaCl (pH 7.4).

![FIG. 2. Poststimulus-time (PST) histograms of an IC neuron obtained before (pre-drug, left) and after (bicuculline, right) bicuculline application.](http://jn.physiology.org/)

Note that this neuron discharged impulses to 25 pps (i.e., the highest 100% pulse-locking repetition rate (PLRR)). At 40 pps, it only responded to the 1st pulse of the PAM train both before and after bicuculline application. This neuron discharged maximally to the 25-pps pulse train (+) before and after bicuculline application. Bicuculline application did not change the neuron’s highest 100% PLRR. Best frequency (BF; kHz) and MT (dB SPL) of the neuron were 21.0 and 36.
and the same current used for bicuculline application was passed through the balanced barrel. Stimulus artifacts were considered negligible when the number of impulses of a neuron was affected <10% after current application (Ebert and Ostwald 1995). Otherwise, the data were discarded, and a new electrode was used for the experiment. Data also were discarded when the impedance of the bicuculline-filled electrode varied >20 MΩ before and after the recording, if the tip of the three-barrel electrode broke when withdrawn from the recording site or when the tips of the single- and the three-barrel electrode separated from each other.

Recorded action potentials were amplified with conventional techniques and sent to a computer (Gateway 2000) for acquisition of the poststimulus time (PST) histogram and the number of impulses of a neuron’s responses over 32 pulse trains. The PST histogram and dot-raster pattern describe the neuron’s temporal response to each pulse train. The number of impulses discharged by a neuron to each pulse and the total number of impulses discharged to each pulse train were both obtained.

To determine the response transfer functions to modulation rate, data were analyzed by plotting the number of impulse against the PRR to derive discharge rate MTFs. Two types of rate MTFs were obtained by plotting the average number of impulse per individual pulse of the train against the PRR and by plotting the total number of impulses per 32 trials of pulse trains against the PRR. For convenience, these two types of rate MTFs are called average rate MTF and total rate MTF. The sync MTFs also were derived by plotting the synchronization coefficients against the PRR. Synchronization coefficients were calculated according to the method modified after Goldberg and Brown (1969) and Rose and Capranica (1985). The synchronization coefficient ranges from zero (no synchronization) to unity (perfect synchronization). The Rayleigh test was used to determine the significance of synchronization coefficients at 0.001 significance level (Buunen and Rhode 1978). The null hypothesis was a uniform distribution of events (no-time locked). A likelihood value ($L$) was determined by $L = 2N(SC)^2$ where $N$ is the number of impulses. For $L > 13.8$, the null hypothesis was rejected.

RESULTS

A total of 66 IC neurons were isolated at depths of 361–1,973 μm. They either discharged one to two impulses (phasic on responders, $n = 41, 62\%$), three to eight impulses (phasic bursters, $n = 19, 29\%$), or many impulses throughout the entire duration of the stimulus (tonic responders, $n = 6, 9\%$), as occurred in previous work (Lu et al. 1997). Response latencies, BFs, and MTs were between 3.5 and 18.5 ms, between 14.2 and 86.3 kHz, and 4–73 dB SPL, respectively, with most <10 ms, between 20 and 70 kHz, and <40 dB SPL, respectively. Whereas 50 neurons responded vigorously to FM pulses, the remaining responded poorly or not at all to FM pulses. These findings are similar to those reported in previous studies on the same species of bat (Caseday and Covey 1992; Jen and Schlegel 1982; Jen et al.
and the number of pulses they could follow (Figs. 2–4). For convenience, a neuron’s ability to respond to presented pulses within a pulse train is defined as the pulse-locking ability. When a neuron discharges impulses to each pulse within a pulse train at a specific PRR, it has 100% pulse-locking ability, and the PRR is defined as 100% pulse-locking repetition rate (PLRR). The highest PRR at which a neuron has 100% pulse-locking ability is defined as the highest 100% PLRR.

Bicuculline application affected the responses of 66 IC neurons to PAM pulse trains in the following four ways.

First, whereas bicuculline application increased the number of impulses of 18 (27%) neurons in response to each presented pulse and the entire PAM pulse train, it did not increase pulse-locking ability. For example, one neuron had its highest 100% PLRR at 25 pps, and it only discharged impulses to the first pulse of the pulse train at 40 pps. Bicuculline application increased the number of impulses in response to each pulse and the total number of impulses to each pulse train (Fig. 2, right vs. left). However, the neuron’s highest 100% PLRR remained at 25 pps, and it only discharged impulses to the first pulse of the pulse train at 40 pps. Second, in addition to increasing the number of impulses of IC neurons in response to each presented pulse, bicuculline application increased the pulse-locking ability of 27 (41%) neurons such that they discharged impulses to more pulses within each pulse train at higher PRRs. However, the application did not change their highest 100% PLRRs. For example, one neuron only discharged impulses to the first pulse of pulse trains at 67, 83, and 100 pps before bicuculline

Bicuculline application affects a neuron’s ability to respond to PAM pulse trains

When stimulated with PAM pulse trains, IC neurons differed in the number of impulses in response to each presented pulse and the number of pulses they could follow (Figs. 2–4). For convenience, a neuron’s ability to respond to presented pulses within a pulse train is defined as the pulse-locking ability. When a neuron discharges impulses to each pulse within a pulse train at a specific PRR, it has 100% pulse-locking ability, and the PRR is defined as 100% pulse-locking repetition rate (PLRR). The highest PRR at which a neuron has 100% pulse-locking ability is defined as the highest 100% PLRR.

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TABLE 2. Effect of bicuculline application on the highest 100% pulse-locking repetition rates (PLRRs) of 66 IC neurons

<table>
<thead>
<tr>
<th>Predrug Highest 100% PLRR (pps)</th>
<th>Postdrug Highest 100% PLRR (pps)*</th>
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</thead>
<tbody>
<tr>
<td>6</td>
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<td>25</td>
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Numbers underlined refer to neurons (45, 68%) the highest 100% PLRRs of which were not changed by bicuculline application. * Number of neurons.

application (Fig. 3, left). Bicuculline application facilitated the neuron in response to more than one pulse within these same pulse trains although the application did not increase the neuron’s highest 100% PLRR (Fig. 3, right). Third, bicuculline application increased the number of impulses and 100% pulse-locking ability of 21 (32%) neurons such that they discharged impulses to each presented pulse at higher PRRs. For example, the highest 100% PLRR of one neuron was increased from 25 pps (Fig. 4, left) to 40 pps on bicuculline application (Fig. 4, right). Table 2 shows the highest 100% PLRR of all 66 IC neurons before and after bicuculline application. It is clear that bicuculline application affected the highest 100% PLRRs of 21 neurons in different ways. Fourth, bicuculline application increased the ability of 62 (94%) neurons to respond to higher PRRs in spite of differences in pulse-locking ability. The highest PRR that these collicular neurons could follow was between 10 and 500 pps (average: 280 ± 50 pps) before bicuculline application. It was between 230 and 500 pps (average: 380 ± 82 pps) after bicuculline application.

In summary, bicuculline application increased the number of impulses of all neurons studied and facilitated their pulse-locking ability in different ways such that they either responded to higher PRRs, responded to more presented pulses or increased the highest 100% PLRRs.

Relationship between response latency and highest 100% PLRRs

Figure 5A shows the distribution of response latencies of all 66 IC neurons in relation to their highest 100% PLRRs obtained before bicuculline application. Response latencies appear to be inversely related to the highest 100% PLRRs. The highest 100% PLRRs tend to be high for neurons with short response latencies and to be low for neurons with long response latencies. Although bicuculline application changed the response latencies of more than half of the collicular neurons and increased the highest 100% PLRRs of about one third of these neurons, it did not change the relationship between response latencies and the highest 100% PLRRs (Fig. 5B).

Bicuculline application on filtering characteristics of rate MTFs

As described above, IC neurons have different pulse-locking abilities so that they discharge different numbers of impulses to each pulse and show different degrees of adaptation to subsequent pulses (Figs. 2–4). Because of this, they discharge maximally to PAM pulse trains at different PRRs and show different filtering characteristics. As described in METHODS, we examined the filtering characteristics of these neurons by plotting their average rate MTF and total rate MTF, which show variation in number of impulses with PRR. Using the same criterion adopted in previous reports (Condon et al. 1991; Feng et al. 1991; Rose and Capranica 1985), a neuron was considered to be tuned to a specific PRR when the number of impulses discharged in response to the most and least preferred PRRs differed by ≥50%.

According to average rate MTFs, all 66 neurons studied showed low-pass filtering characteristics. The average number of impulses discharged to each pulse decreased by >50% with increasing PRR. However, the rate of decrease was different among individual neurons such that all average rate MTFs have different slopes (Fig. 6). According to total rate MTFs, four types of filtering characteristics can be described within 1- to 250-pps PRR tested. 1) Band-pass filters in which the maximal number of impulses at the most preferred repetition rate was ≥50% greater than the number of impulses at the two minimal responses. These neurons had inverted ‘‘V’’-shaped rate MTFs (Fig. 7A). For convenience, the most preferred PRR is defined as the best repetition rate (BRR). BRRs were only identified for 48 IC neurons with band-pass filtering characteristics. 2) Low-pass filters in which the maximal number of impulses discharged at low PRRs was 50% greater than the minimal number of
impulses obtained at high PRRs (Fig. 7B). 3) High-pass filters in which the maximal number of impulses obtained at the high PRRs was 50% greater than the number of impulses obtained at low PRRs (Fig. 7C). 4) All-pass filters in which the number of impulses discharged at all PRRs tested never differed by >50% (Fig. 7D).

We examined the BRR of 48 neurons with band-pass filtering characteristics before and after bicuculline application. Bicuculline application increased the number of impulses of each neuron in response to pulses delivered at each PRR such that the neuron’s postdrug total rate MTF is always higher than the predrug total rate MTF (Fig. 8, Ab, Bb, and Cb vs. Aa, Ba, and Ca). Whereas bicuculline application did not affect the BRRs of 28 neurons (Fig. 8A), the application either increased (n = 11; Fig. 8B) or decreased (n = 9; Fig. 8C) the BRR of the remaining 20 neurons. Table 3 shows the distribution of the BRR of these 48 neurons before and after bicuculline application.

Bicuculline application did not change the average rate MTF-based filtering characteristics. In contrast, bicuculline application changed the total rate MTF-based filtering char-
characteristics of 14 (21%) neurons. Table 4 shows the distribution of total rate MTF-based filtering characteristics of all 66 IC neurons before and after bicuculline application. Whereas the application changed only 4 band-pass neurons into 3 low-pass and 1 all-pass, it changed 10 low-pass neurons into 8 band-pass and 2 all-pass neurons. In spite of these changes, most IC neurons had band-pass filtering characteristics both before and after bicuculline application.

Bicuculline application on filtering characteristics of sync MTFs

To determine the neuron’s selectivity for temporal information, we also examined the filtering characteristics of IC neurons by plotting their sync MTFs to show variation in the synchronization coefficient with PRR. The sync MTF of each neuron was obtained both before and after bicuculline application.

Although bicuculline application affected the filtering characteristic of some IC neurons (see further, Table 5), four types of filtering characteristics in sync MTFs can be described. 1) Low-pass filters in which the synchronization coefficient started at or near 1.0 at lower PRRs and sharply reduced by >50% at high PRRs (Fig. 9A). 2) All-pass filters in which all synchronization coefficients were generally high and never differed from each other by >50% (Fig. 9B). 3) Band-suppression filters in which the synchronization coefficients were high at both low and high PRRs but they dropped by >50% to a minimum at a specific PRR. In other words, the sync MTFs had “V”-shaped functions with skirts at both low and high repetition rates (Fig. 9C). 4) Band-suppression–band-pass filters in which the synchronization coefficient started at or near 1.0 and sharply dropped to a minimum at a certain PRR. It then increased to a new maximum before dropping again to a new minimum. In other words, the sync MTFs of these neurons are tuned to two specific PRRs. The filtering characteristics of these neurons appear to have the combined characteristics of a band-suppression and a band-pass filter (Fig. 9D).

Table 5 shows the distribution of sync MTFs-based filtering characteristics of all 66 IC neurons before and after bicuculline application. Bicuculline application changed the filtering characteristics of 19 (29%) neurons. Whereas the application changed seven all-pass neurons, all seven band-pass neurons and two band-suppression–band-pass neurons into other filtering characteristics, it only changed three low-pass neurons (28, 58%) the BRRs of which were not changed by bicuculline application. Rate MTF, discharge rate-based modulation transfer function. * Number of neurons.

Table 3. Effect of bicuculline application on the best repetition rates (BRRs) of 48 IC neurons with band-pass total rate MTF filtering characteristics

<table>
<thead>
<tr>
<th>Predrug BRR (pps)</th>
<th>Postdrug BRR (pps)*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>4</td>
</tr>
<tr>
<td>4</td>
<td>2</td>
</tr>
<tr>
<td>6</td>
<td>2</td>
</tr>
<tr>
<td>10</td>
<td>3</td>
</tr>
<tr>
<td>25</td>
<td>28</td>
</tr>
<tr>
<td>40</td>
<td>8</td>
</tr>
<tr>
<td>67</td>
<td>3</td>
</tr>
<tr>
<td>83</td>
<td>2</td>
</tr>
</tbody>
</table>

Numbers underlined refer to neurons (28, 58%) the BRRs of which were not changed by bicuculline application. Rate MTF, discharge rate-based modulation transfer function. * Number of neurons.

Table 4. Effect of bicuculline application on total rate MTF filtering characteristics of 66 IC neurons

<table>
<thead>
<tr>
<th>Predrug</th>
<th>Postdrug*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Band-Pass</td>
</tr>
<tr>
<td>Band-pass</td>
<td>52 (79%)</td>
</tr>
<tr>
<td>Low-pass</td>
<td>12 (18%)</td>
</tr>
<tr>
<td>High-pass</td>
<td>2 (3%)</td>
</tr>
<tr>
<td>Total</td>
<td>56 (85%)</td>
</tr>
</tbody>
</table>

Numbers underlined refer to neurons (52, 79%) the rate MTF filtering characteristics of which were not changed by bicuculline application. Percentage of neurons are in parentheses; * number of neurons.
TABLE 5. Effect of bicuculline application on the sync MTF filtering characteristics of 66 IC neurons

<table>
<thead>
<tr>
<th></th>
<th>Predrug</th>
<th>Postdrug*</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>n</td>
<td>Low-Pass</td>
</tr>
<tr>
<td>Low-pass</td>
<td>41 (62%)</td>
<td>38</td>
</tr>
<tr>
<td>All-pass</td>
<td>11 (17%)</td>
<td>3</td>
</tr>
<tr>
<td>BS</td>
<td>7 (10.5%)</td>
<td>4</td>
</tr>
<tr>
<td>BSBP</td>
<td>7 (10.5%)</td>
<td>2</td>
</tr>
<tr>
<td>Total</td>
<td>47 (71%)</td>
<td>4 (6%)</td>
</tr>
</tbody>
</table>

Numbers underlined refer to neurons (47, 71%) the synchronization coefficient-based modulation transfer function (sync MTF) filtering characteristics of which were not changed by bicuculline application. Percentage of neurons are in parentheses. BS, band-suppression; BSBP, band-suppression–band-pass. * Number of neurons.

pass neurons into three band-suppression–band-pass neurons. In spite of these changes, most IC neurons have low-pass filtering characteristics in sync MTFs both before and after bicuculline application. Figure 10, A and B, shows the overall shape of eight representative low-pass sync MTFs that were not affected by bicuculline application. It is clear that sync MTFs-based filtering characteristics obtained before and after bicuculline application are extremely similar.

DISCUSSION

Number of impulses and PRRs

In this study, we have demonstrated that IC neurons differed in pulse-locking ability and in the number of impulses in response to each presented pulse of PAM pulse trains. Individual IC neurons discharged impulses to all, some, or only the first pulse of presented PAM pulse trains (Figs. 2–4). Because of this, few IC neurons discharged maximally to the PAM pulse train with the largest number of pulses (i.e., the highest PRR). Most neurons simply adapted to different degrees according to their pulse-locking ability and showed band-pass total rate MTFs (Fig. 7A, Table 4). Similar findings have been reported for IC neurons of cats (Langner and Schreiner 1988) and bats (Condon et al. 1994; Pinheiro et al. 1991).

The present finding is different from a previous study on the auditory midbrain, the torus semicircularis, of the northern leopard frog (Gooler and Feng 1992). That study reported that in response to PAM pulse trains with 10-ms pulses delivered between 3.33 and 100 pps, most neurons (64%) gave greater spike counts with increasing amplitude-modulated (AM) rate, yielding high-pass rate MTFs. This discrepancy is likely due to the fact that within this range of PRRs, most neurons have high pulse-locking ability and can follow all or most presented pulses within each pulse train. If short pulse duration and AM rates >100 pps were incorporated in the PAM pulse trains, neurons likely would not always give a greater number of impulses to pulse trains at higher AM rates because of deterioration in pulse-locking ability. Then most neurons would have band-pass rate MTFs instead of high-pass rate MTFs. A similar argument has been presented in a previous study (Feng et al. 1991).

Bicuculline application affected the pulse-locking ability of IC neurons

The present study shows that bicuculline application increased the number of impulses of IC neurons in response to each presented pulse. The application also increased the pulse-locking ability of IC neurons such that they could respond to higher PRRs (Figs. 2–4). We believe that increased neuronal excitability by bicuculline application is primarily due to the change in the recovery cycles of these IC neurons.

The recovery cycle is an important neuronal property that

FIG. 9. Representative synchronization coefficient-based modulation transfer function (sync MTF) curves of 4 IC neurons showing the low-pass (A), all-pass (B), band-suppression (C), and band-suppression–band-pass (D) filtering characteristics. Ordinates and abscissae represent the synchronization coefficient and pulse repetition rate in pps. ---, half value of unity, i.e., 0.5. ●, insignificant synchronization coefficients (P > 0.001; 3 points in A, 1 in C, and 2 in D). BF (kHz) and MT (dB SPL) of these neurons were A: 22.0, 37; B: 21.0, 36; C: 30.0, 23; and D: 15.4, 34.
determines a neuron’s ability to respond to sound pulses delivered at different PRRs. A neuron’s recovery cycle is determined primarily by the refractory period after excitation and inhibitory inputs from other neurons. Because IC neurons differ in recovery cycles, they differ in pulse-locking ability to respond to PAM pulse trains. When the PRR of a PAM pulse train is increased, the interpulse interval is shortened and this prevents a neuron from recovering completely from previous stimulation. At higher PRRs, the neuron would completely cease responding due to the extremely short interpulse interval. Previous studies showed that most IC neurons receive GABAergic inhibition (Casseday et al. 1994; Fubara et al. 1996; Johnson 1993; Park and Pollak 1993a,b; Vater et al. 1992). It also has been shown that GABAergic disinhibition by bicuculline application increased the excitability and shortened the recovery cycles of most IC neurons (Ebert and Ostwald 1995; Faingold et al. 1989, 1991; Lu et al. 1997; Palombi and Caspary 1992; Park and Pollak 1993a,b; Vater et al. 1992; Yang et al. 1992). All these studies suggest that our observation of increased pulse-locking ability and increased responses to higher PRRs of IC neurons on bicuculline application are likely due to increased neuronal excitability and shortening of the recovery cycles on GABAergic disinhibition.

IC neurons often responded only to the first or the initial few pulses in a PAM pulse train at certain PRRs (e.g., Fig. 2: 40 pps; Fig. 3: 67, 83, and 100 pps; Fig. 4: 67 pps). We believe that this observation is primarily due to slow recovery cycles of these neurons. This change in temporal selectivity with PRR has been reported in a study of auditory cortical neurons of bats (Wong et al. 1992).

The increased pulse-locking ability of most IC neurons to respond to PAM pulse trains and the increased number of neurons with band-pass filtering characteristics in total rate MTFs after bicuculline application (Figs. 2–4, Table 4) certainly increase the dynamic aspects of auditory temporal processing. While this dynamic change in temporal selectivity may be significant for auditory signal processing in general, it is particularly important for echolocation in bats, which use temporal information carried by echoes to detect and recognize their prey (Griffin 1958).

If a bat can use this mechanism of changing dynamic aspect of temporal selectivity during hunting, it certainly would enhance its versatility in response to PRRs, which change systematically during different phases of hunting. Recent studies show that corticofugal pathway may regulate the directional sensitivity, frequency tuning, intensity coding, and delay tuning of IC neurons (Sun et al. 1989, 1996; Yan and Suga 1996). It is likely that a bat also may use this pathway to regulate the temporal selectivity during auditory information processing.

**Relationship between latency and BRRs and 100% pulse-locking ability**

As shown in previous studies in cats (Langner and Schreiner 1988; Langner et al. 1987) and bats (Pinheiro et al. 1991), the BRRs of 48 IC neurons with band-pass total rate MTFs obtained both before and after bicuculline application were correlated significantly with their response latencies (Fig. 11, A vs. B). However, BRR is not correlated with BF of these neurons (Fig. 12). A previous study showed a similar discrepancy between BRR (which represents temporal selectivity) and BF (which represents frequency selectivity) (Pinheiro et al. 1991).

We have demonstrated that the highest 100% PLRRs of
IC neurons were correlated significantly with the response latencies regardless of bicuculline application (Fig. 5). This finding and the correlation between response latencies and BRRs (Fig. 11) suggest that IC neurons with high highest 100% PLRRs have high BRRs. Indeed, a linear analysis of the distribution of BRRs and highest 100% PLRRs produced a high correlation coefficient of 0.78 (n = 48, P < 0.0001).

Comparisons of rate MTFs and sync MTFs with previous work

To determine a neuron’s selectivity for temporal information, previous studies determined the filtering characteristics of auditory neurons by deriving sync MTFs and rate MTFs. Whereas the sync MTFs were derived by plotting the synchronization coefficients against the PRRs, the rate MTFs were derived by plotting the normalized response or percent maximum response (Condon et al. 1994, 1996; Feng et al. 1991; Gooler and Feng 1992; Hall and Feng 1991) or the discharge rate (spikes/s) (Rose and Capranica 1985) against PRRs. This is similar to our total rate MTF in which the total number of impulses obtained from each pulse train was plotted against PRRs to obtain rate MTFs. We also use the average number of impulse per individual pulse of the train to construct the average rate MTFs. Using these two rate MTFs and sync MTFs, we determine the effect of bicuculline application on the filtering characteristics of each neuron.

In a previous study (Rose and Capranica 1985) on the midbrain torus semicircularis of leopard frogs (Rana pipiens), the rate MTFs of midbrain neurons were classified into five types, AM nonselective (35%), AM high-pass (9%), AM low-pass (17%), AM band-suppression (9%), and AM-tuned (30%). Other studies on the midbrain neurons of frogs show similar patterns (Epping and Eggermont 1986; Walkowiak 1984). However, all five classes of neurons showed high synchronization coefficients at low AM rates and low synchronization coefficients at high AM rates (i.e., low-pass characteristics in sync MTFs). Thus when rate MTFs were compared with sync MTFs, the temporal selectivity of the frog’s midbrain neurons was not evident from their synchronization characteristics. A comparable study in the dorsal medullary nucleus of the leopard frog showed that 27% of the neurons had band-pass filtering characteristics in rate MTFs but 66% had low-pass filtering characteristics in sync MTFs (Hall and Feng 1991).

A similar study of the responses of IC neurons to temporally patterned sound pulses has been conducted in the little brown bat, Myotis lucifugus (Condon et al. 1994). In this study, unmodulated trains of tone pulses with different PRRs (TPu) were used to examine the ability of IC neurons to follow pulses with different PRRs. The TPu was characterized by setting pulse duration at 2 ms (0.5-ms rise-decay times), intensity at 10 dB above MT, train duration at 400 ms, carrier pulse at BF or downward 80 to 20 kHz sweep, and PRR varying from 5 to 400 pps. The filtering characteristics of 130 IC neurons showed a large number of band-pass (56%) and low-pass (22%) in rate MTFs, but most (75%) IC neurons showed low-pass filtering characteristics in sync MTFs. Our study also showed that a large number of band-pass (79%) and low-pass (18%) neurons in total rate MTFs (Table 4) but most (62%) neurons had low-pass filtering characteristics in sync MTFs (Table 5).

All of these comparisons show that auditory neurons in frogs and bats have a great deal of selectivity in temporal signals as demonstrated by the different types of filtering characteristics of total rate MTFs. However, the temporal selectivity represented by total rate MTFs is not evident in their sync MTFs because most neurons show low-pass characteristics in their sync MTFs. On the other hand, in consistent with our previous study (Pinheiro et al. 1991), we found that all 66 neurons showed low-pass filtering characteristics in average rate MTFs on bicuculline application. Thus it appears that filtering characteristics of neurons determined in average rate MTFs rather than total rate MTFs are more comparable with sync MTFs.

In summary, our results showed that bicuculline application increased the number of impulses, changed discharge patterns and response latency of most IC neurons (Figs. 2–4). Temporal selectivity of most neurons in average rate MTFs, total rate MTFs, and sync MTFs of most neurons are not affected by bicuculline application (Tables 4 and 5). These observations suggest that GABAergic inhibition may regulate effectively in the firing rate of auditory neurons but may not be as effective in regulating the temporal information processing of the neurons.
REFERENCES

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