Signal-to-noise ratio in the membrane potential of the owl’s auditory coincidence detectors

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Ashida G, Funabiki K, Kuokkanen PT, Kempter R, Carr CE. Signal-to-noise ratio in the membrane potential of the owl’s auditory coincidence detectors. J Neurophysiol 108: 2837–2845, 2012. First published August 29, 2012; doi:10.1152/jn.00366.2012.—Owls use interaural time differences (ITDs) to locate a sound source. They compute ITD in a specialized neural circuit that consists of axonal delay lines from the cochlear nucleus magnocellularis (NM) and coincidence detectors in the nucleus laminaris (NL). Recent physiological recordings have shown that tonal stimuli induce oscillatory membrane potentials in NL neurons (Funabiki K, Ashida G, Konishi M. J Neurosci 31: 15245–15256, 2011). The amplitude of these oscillations varies with ITD and is strongly correlated to the firing rate. The oscillation, termed the sound analog potential, has the same frequency as the stimulus tone and is presumed to originate from phase-locked synaptic inputs from NM fibers. To investigate how these oscillatory membrane potentials are generated, we applied recently developed signal-to-noise ratio (SNR) analysis techniques (Kuokkanen PT, Wagner H, Ashida G, Carr CE, Kempter R. J Neurophysiol 104: 2274–2290, 2010) to the intracellular waveforms obtained in vivo. Our theoretical prediction of the band-limited SNRs agreed with experimental data for mid- to high-frequency (>2 kHz) NL neurons. For low-frequency (≤2 kHz) NL neurons, however, measured SNRs were lower than theoretical predictions. These results suggest that the number of independent NM fibers converging onto each NL neuron and/or the population-averaged degree of phase-locking of the NM fibers could be significantly smaller in the low-frequency NL region than estimated for higher best-frequency NL.

Signal-to-noise ratio; owl
MATERIALS AND METHODS

Electrophysiology. In this article we use the electrophysiological data described in Funabiki et al. (2011), where experimental procedures were described in detail. Animal husbandry and experimental protocols conform to National Institutes of Health Guidelines for the Care and Use of Laboratory Animals and were approved by the Animal Care and Use Committee of the California Institute of Technology. Nine barn owls (Tyto alba) of both sexes were anesthetized by intramuscular injection of ketamine (25 mg/kg) and diazepam (1.3 mg/kg) and placed in a sound-attenuating chamber. Additional doses of ketamine were administered to maintain a suitable level of anesthesia. Recordings were made with specially designed coaxial glass electrodes (>80 MΩ; see Funabiki et al. 2011 for detailed configuration and operations).

Custom-written software (xdphys; California Institute of Technology) together with the TDT2 signal-processing system (Tucker Davis Technology, Gainesville, FL) was used for controlling acoustic stimuli and collecting data. Acoustic stimuli were passed through a digital-to-analog converter, filtered, attenuated, impedance-matched, and delivered to the animal via custom-made earphones placed into the ear canals. Sound pressure levels were calibrated before recordings using built-in miniature microphones.

Voltage and current data were recorded with Neurodata IR-183 (Cygnus Technology, Delaware Water Gap, PA) or Axoclamp 2A amplifier in bridge mode (Axon Instruments, Union City, CA). Responses to acoustic stimuli were continuously monitored until the electrode reached the NL in the brain stem. When a single unit was obtained intracellularly, we measured the DC drop at penetration and the resting potential without holding current. Data from neurons with resting potentials below −50 mV were considered as successful intracellular recordings and used for further analyses. Intracellular membrane potential waveforms were digitized and stored at a sampling rate of 48,077 Hz.

The sound stimulus consisted of a tone burst of 60 or 100 ms in duration with a 3-ms rise/fall time. The average binaural intensity was fixed to 40 dB SPL. The best frequency (BF) of each neuron was estimated audiovisually by determining the stimulus frequency that evoked the strongest response, and the BF was confirmed by measuring the periodicity of ITD tuning curves collected using broadband noise. Note that in the physiological ITD range, an ITD tuning curve with broadband noise is practically indistinguishable from an ITD tuning curve with a tone at the BF (Fischer et al. 2008). To measure the ITD-dependent spike rate change, the ITD of the stimulus tone at

Fig. 1. Schematic drawings of the synaptic input and the signal-to-noise ratio (SNR) of the nucleus laminaris (NL) neuron. A: formation of the oscillatory synaptic input. A tonal stimulus induces periodic spike rate changes of cochlear nucleus magnocellularis (NM) neurons. Phase-locked spikes of NM fibers that converge onto an NL neuron create a periodically oscillating synaptic input. Note that troughs of the sound-driven input rate can be below the baseline because of the high spontaneous spike rate of NM fibers (see Funabiki et al. 2011 for related discussion). EPSC, excitatory postsynaptic current. B: summation of the synaptic input from ipsi- and contralateral NM fibers. The oscillation amplitude of the bilateral synaptic input to NL is maximal when the 2 inputs arrive perfectly in phase, whereas it is minimal when the 2 inputs are out of phase. Note that, for clarity, higher harmonics and noise components are not included in this schematic. δ, Phase difference. C: power spectral densities (PSDs) of the input and the membrane potential. Top, PSD of the phase-locked NM inputs. Phase-locking produces the fundamental frequency component and its harmonics in addition to the flat baseline noise. Middle, PSD of the total synaptic input. The low-pass property of the synaptic filtering leads to the PSD of the synaptic input decaying with frequency. Bottom, PSD of the membrane potential. Synaptic input is further filtered by the low-pass membrane processes. Because of the synaptic and membrane filtering, the second harmonic is, in general, smaller than the fundamental frequency component by a few orders of magnitude. Whereas the synaptic and membrane filters alter the overall shape of the PSDs, the band-limited SNR (indicated by gray arrows) remains unchanged throughout these processes.
BF was varied in steps of either one-tenth of the period of the tone or 30 μs. Tonal stimuli were repeated typically three times for each ITD.

**Analysis of the sound analog potential.** Sound analog potentials were analyzed according to the methods we provided previously (Funabiki et al. 2011). Custom-written Matlab (The MathWorks, Natick, MA) scripts were used for all data analyses. From the stored intracellular traces, spikes were discriminated by a voltage threshold to compute firing rate. The membrane potential of an NL neuron generally oscillated at the stimulus frequency during tonal stimulation (e.g., Fig. 2, A and E). We quantified this oscillation by fitting the membrane potential waveform during the stimulus with a cosine function: \( y(t) = A_0 + B_0 \cos(2\pi f_t + \phi) \), where \( A_0 \) is the average potential, \( B_0 \) is the oscillation amplitude, \( f_t \) is the stimulus frequency, \( t \) is time, and \( \phi \) is the phase shift. The cosine fitting was performed to unfiltered traces (see Fig. 2). Note that cosine fitting to the bandpass-filtered traces leads to the same result because filtering does not change the Fourier component at the stimulus frequency. The amplitude \( B_0 \) of the fitting function was considered as the “AC” component of the membrane potential. An AC-ITD curve (e.g., Fig. 2, D and H) was obtained by fitting with an absolute cosine function: \( B_{f}(x) = B_1 \left[ \cos(\pi f_t (x - x_0)) \right] \), with \( x_0 \) being ITD. The AC amplitude \( B_{f}(x) \) achieves its maximum \( B_1 \) when \( x = x_0 \). The absolute cosine function is derived from the sum of two sine functions corresponding to the two monaural inputs (see Ashida et al. 2007 for more discussion).

**SNR analysis.** Membrane potential waveforms between 10 and 50 ms after the stimulus onset were used in our frequency analyses. The waveforms were resampled at 51,200 Hz (yielding 2,048 points) by linear interpolation to obtain integer numbers of samples in each 40-ms segment and were then Fourier-transformed. The frequency resolution of the Fourier transform was 25 Hz. Note that the stimulus frequency we used was always a multiple of 25 Hz. The power spectral density (PSD) is the square of the absolute value of the Fourier transform.

The band-limited SNR is defined as the ratio of the PSD at the stimulus frequency \( f_s \) to that at the surrounding frequencies \( f \neq f_s \) (e.g., Fig. 3, A and D; see Kuokkanen et al. 2010 for details). Namely,

\[
\text{SNR}(f_s) = \frac{\text{PSD}(f_s)}{\text{PSD}(f)} \quad \text{for } f \sim f_s \text{ but } f \neq f_s
\]

Figure 1C shows a schematic example of the band-limited SNR. The PSD of the membrane potential generated by phase-locked synaptic inputs shows peaks at the stimulus frequency and its harmonics. It should be noted that the definition of SNR may vary between studies (e.g., Englitz et al. 2009; Svirskis et al. 2002). In the current study, the band-limited SNR is the peak height of the PSD measured from the baseline noise level (gray arrows in Fig. 1C). The absolute values and the overall shapes of the PSDs may differ due to synaptic and membrane filtering (3 curves in Fig. 1C). Effects of these filters, however, are canceled during the calculation of the SNR (see **Theoretical estimation of SNR** for more theoretical background), and thus the relative heights of the peaks remain unchanged (gray arrows in Fig. 1C). Furthermore, in recordings in vivo, the frequency characteristics of the recording apparatus may also affect the shape of the PSD curve. The band-limited SNR, however, is not affected by the frequency profile of the recording system, and the SNR was proven to be useful in characterizing the extracellular field potential in the owl’s NL (Kuokkanen et al. 2010).

To estimate the noise level (denominator of Eq. 1) from our physiological data, we averaged the PSD amplitudes over the 1-kHz interval between \( f_s - 0.5 \text{ kHz} \) and \( f_s + 0.5 \text{ kHz} \), excluding the peak at the stimulus frequency \( f_s \). This bandwidth, in general, has to be determined to include a sufficient number of data points in the estimation. However, the frequency band should not be excessively wide, because the overall shape of the PSD curve may skew the estimate. As a compromise, we chose a 1-kHz band for the calculation of the band-limited SNR, as we did in our previous study (Kuokkanen et al. 2010). See **Appendix** for more discussion.

**Theoretical estimation of SNR.** Kuokkanen et al. (2010) derived the analytical expression for the PSD:

\[
\text{PSD}(f_s) = M^2 \lambda^2 \left( K(f_s) \right)^2 T, \quad \text{for } f \neq f_s
\]

where \( M \) is the number of presynaptic NM fibers, \( \lambda \) is the average rate of firing of these fibers, \( r \) is their average degree of phase locking (vector strength; Goldberg and Brown 1969), and \( T \) is the length of the analyzed time window (fixed to 40 ms in this study). The symbol \( K(f) \) denotes the Fourier transform of the potential waveform associated with each NM spike. We assumed the activities of the NM fibers to be statistically independent from each other, and the unitary input from each NM fiber to have an identical waveform. From Eqs. 1–3, we now have the theoretical expression for the band-limited SNR:

\[
\text{SNR}(f_s) = M \lambda^2 r^2 T.
\]

Note that the effect of the unitary synaptic input described by \( K(f) \) has been canceled. Only the presynaptic factors \( M, \lambda, \) and \( r \) appear in the equation, enabling us to focus on these parameters. This property of \( Eq. 4 \) is a major advantage of using the band-limited SNR.

To calculate theoretical SNRs from \( Eq. 4 \), we used previously reported parameter values (Table 1). The upper limits (lower limits or medians) for these parameters were used to calculate the theoretical upper limit (lower limit or median) for the SNR. Note that the number and the spike rate of NM fibers were assumed to be frequency independent because there is no report that systematically investigated their frequency dependence (see also **RESULTS AND DISCUSSION**). We used the vector strengths of individual NM fibers as a simple estimate of population vector strength.

**RESULTS AND DISCUSSION**

**In vivo intracellular recordings from the owl’s NL.** NL neurons are coincidence detectors that change their output spike rate by sensing the time disparities between the ipsilateral and contralateral inputs (Carr and Konishi 1990). In vivo intracellular recordings from NL neurons revealed that tonal stimuli induce sinusoidal oscillations in the membrane potential (Fig. 2, A and E), termed the sound analog potential (Funabiki et al. 2011). These oscillation frequencies matched the stimulus tone frequencies (see also the PSDs in Fig. 3, A and D), and they resemble oscillatory membrane potentials found in auditory hair cells (Russell and Sellick 1978).

As reported by Funabiki et al. (2011), the amplitude of the membrane potential oscillation changed with ITD (Fig. 2, D and H), as did the output spike rate of the neuron (Fig. 2, C and G). Small spikes (typically ~10-mV amplitude) suggest that they are generated at a remote site, perhaps at the first node of Ranvier, and also suggest that the cell body may not be excitable (Ashida et al. 2007; Kuba et al. 2006). Small spikes due to the segregation of synaptic input and spike generation also characterize the mammalian auditory coincidence detectors of the medial superior olive (MSO) (Scott et al. 2005) and the octopus cells of the posteroventral cochlear nucleus (Golden et al. 1999).

The generation of the sound analog membrane potential in the NL neuron can be explained by converging phase-locked synaptic inputs (Ashida et al. 2007; Kempter et al. 1998; Slee et al. 2010). In principle, phase-locked spike sequences from the NM axons are filtered by synaptic and membrane processes, leading to oscillatory membrane potentials in NL (Fig.
Fig. 2. In vivo intracellular recordings from barn owl NL. Two representative examples are shown [A–D: best frequency (BF) = 3,400 Hz; E–H: BF = 2,000 Hz]. A: membrane potential of an NL neuron. Unfiltered and bandpass-filtered (3,350–3,450 Hz) traces are shown. A binaural tonal stimulus at BF is delivered at a favorable interaural time difference (ITD; ±2 μs). The membrane potential oscillates at the same fundamental frequency as the stimulus tone. B: membrane potential of the same NL neuron as in A, but for a binaural tonal stimulus at BF delivered at an unfavorable ITD (−109 μs). The oscillation amplitude of the membrane potential is much smaller than that with a favorable ITD. Arrowheads in A and B indicate spikes. C: ITD-dependent spike rate of the NL neuron. Error bars are SD. D: ITD-dependent oscillation amplitude (AC) of the membrane potential. The solid line shows the absolute cosine fit (see MATERIALS AND METHODS). E: membrane potential of another NL neuron. Unfiltered and bandpass-filtered (1,950–2,050 Hz) traces are shown. The binaural tonal stimulus at BF is delivered at a favorable ITD (±193 μs). F: membrane potential of the same neuron as in E, but for a binaural tonal stimulus at BF delivered at an unfavorable ITD (−59 μs). G: ITD-dependent spike rate. Error bars are SD. H: ITD-dependent membrane AC component. The solid line shows the absolute cosine fit. To calculate the spike rates (C and G) and oscillation amplitudes (D and H), three 40-ms trials per ITD were used.
The phase differences between inputs from ipsi- and contralateral NM, which are the consequence of ITD, lead to the periodic changes in the AC component of the binaural input (Fig. 1B). In the following sections, we examine, using the band-limited SNR, how the sound analog potential in NL is affected by various input parameters of converging NM fibers.

**Band-limited SNR of NL neurons.** The PSDs of the NL membrane potential revealed the frequency profile of the sound analog potential, including the fundamental frequency component and its harmonics (peaks in Fig. 3, A and D), as well as noise components that decay with frequency (Fig. 3, A, B, D, and E). Note that the higher harmonics were smaller than the main signal component by several orders of magnitude (Fig. 3, A and D). The overall shape of the PSD curve should reflect not only the properties of the presynaptic NM fibers but also those of the synaptic and membrane filters (Fig. 1C). To exclude the effect of these filters, we introduced the band-limited SNR, defined as the ratio of the signal PSD to the PSD at neighboring frequencies (Fig. 1C; see MATERIALS AND METHODS). The band-limited SNR was also useful in characterizing the extracellular field potential of owl’s NL because the SNR ranged over three orders of magnitude, whereas the signal and noise levels varied over seven and four orders of magnitude, respectively (Kuokkanen et al. 2010).

**Table 1. Parameter values for the theoretical estimation of the band-limited SNR**

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Value</th>
<th>Reference</th>
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<tbody>
<tr>
<td>Mean spike rate of each NM fiber ( \lambda_0 )</td>
<td>Upper limit: 500 Hz; Median: 400 Hz; Lower limit: 300 Hz</td>
<td>Peña et al. 1996</td>
</tr>
<tr>
<td></td>
<td>Upper limit: 300 fibers; Median: 200 fibers; Lower limit: 100 fibers</td>
<td>Carr and Boudreau 1993</td>
</tr>
<tr>
<td>Vector strength of phase-locked NM spiking ( r )</td>
<td>Upper limit: 0.84–0.18 ln(f); Median: 0.76–0.24 ln(f); Lower limit: 0.64–0.30 ln(f)</td>
<td>Köppl 1997a</td>
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Equations for the vector strength were derived from a linear fitting of the existing data; \( f \) is frequency in kHz. SNR, signal-to-noise ratio; NM, nucleus magnocellularis; NL, nucleus laminaris.

Fig. 3. PSD and band-limited SNR of NL neurons. Data are from the same 2 neurons as in Fig. 2 (A–C: BF = 3,400 Hz; D–F: BF = 2,000 Hz), A: PSD of the membrane potential trace with a favorable ITD. The curve shows a sharp peak at the stimulus frequency (3,400 Hz, open arrowhead) and a smaller peak at the second harmonic (6,800 Hz; filled arrowhead). Dashed gray lines show the stimulus frequency component (“signal”) and the baseline “noise” at the surrounding frequencies. The height of the signal above the noise gives the band-limited SNR. B: PSD of the membrane potential trace with an unfavorable ITD. The peak at the stimulus frequency (open arrowhead) is much smaller than that in A. C: ITD dependence of band-limited SNR. Note the logarithmic scale in the ordinate. D: PSD of the trace with a favorable ITD. The stimulus frequency component (2,000 Hz) is indicated by an open arrowhead, and higher harmonics are indicated by filled arrowheads. E: PSD of the trace with an unfavorable ITD. F: band-limited SNR of the same neuron. The solid lines in C and F show absolute cosine fits (see MATERIALS AND METHODS). Three 40-ms traces per ITD were used in C and F. To reduce jitter in the PSD curves (in A, B, D, and E), we averaged 3 PSDs at each ITD.
Figure 3, C and F, shows the band-limited SNRs for the NL responses depicted in Fig. 2. Similarly to the AC amplitude (Fig. 2, D and H), the ITD dependence of the band-limited SNR can be predicted by an absolute cosine function (solid lines in Fig. 3, C and F; see MATERIALS AND METHODS). The peak and trough positions of the spike rate (Fig. 2, C and G), the AC amplitude (Fig. 2, D and F), and the SNR (Fig. 3, C and F) coincided well. Maximum band-limited SNRs of the two neurons shown were 28.5 dB (Fig. 3C; BF = 3,400 Hz) and 20.5 dB (Fig. 3F; BF = 2,000 Hz). The maximum SNR is expected to be attained when inputs from ipsi- and contralateral NM fibers arrive at the NL neuron with a zero delay or with a phase delay of an integer multiple of 2π (“in phase” in Fig. 1B). The maximum band-limited SNRs of NL neurons (BF = 0.8–5.6 kHz) ranged between 20 and 30 dB (Fig. 4). Next, we compare these SNR values with our theoretical predictions.

**Theoretical prediction of band-limited SNR.** The band-limited SNR is related to the number \( M \) of NM fibers, the mean spike rate \( \lambda_0 \) of these fibers, and their average vector strength \( r \). In MATERIALS AND METHODS, we showed under what conditions the relation SNR = \( M\lambda_0r^2T \) holds, with \( T \) being the analyzed time length, fixed to 40 ms in this study. Known parameter ranges (Table 1) were used to calculate the upper and lower bounds and the median value for SNR. The degree of phase locking measured by the vector strength gradually decreased with frequency (Köppl 1997a). The number of NM inputs and their mean spike rate, however, were assumed to be frequency independent because only non-frequency-specific data are available (Carr and Boudreau 1993; Peña et al. 1996).

Our theoretical estimates of the band-limited SNR for a 3.5-kHz NL neuron ranged from 19.2 to 33.6 dB, which matched well with the SNRs observed with 3- to 4-kHz NL neurons (Fig. 4). For all NL neurons with a BF of 3 kHz and above, the SNRs calculated from the in vivo data lay within the theoretical upper and lower bounds (Fig. 4). The agreement between the theory and experiment further supports the presumption that the oscillatory membrane potentials in NL are generated by phase-locked excitatory synaptic input from NM fibers (Ashida et al. 2007; Funabiki et al. 2011).

In contrast to 3- to 4-kHz neurons, NL neurons with a BF of 2 kHz or below showed SNRs 5–10 dB lower than the theoretical lower bounds (Fig. 4), although better phase locking in low-BF cells (Köppl 1997a) should lead to higher SNRs. These low-frequency neurons (≥2 kHz) have significantly different SNRs from mid-to-high-frequency (>2 kHz) NL neurons (low BF: 22.5 ± 2.23 dB, \( n = 5 \); mid to high BF: 26.2 ± 2.78 dB, \( n = 16 \); means ± SD, \( P = 0.014 \), unpaired t-test). These results are consistent with previous studies that demonstrated anatomical (Köppl and Carr 1997) and physiological differences (Carr and Köppl 2004; Funabiki et al. 2011) between low- and mid-to-high-frequency NL neurons. Next, we reexamine the assumptions of the model and discuss possible reasons for the lower SNRs in low-BF neurons.

**Model assumptions.** For the derivation of the equation SNR = \( M\lambda_0r^2T \), we assumed that each NL neuron received converging inputs from 1) \( M \) statistically independent fibers, 2) spiking at the mean spiking rate \( \lambda_0 \), with 3) a population-averaged vector strength \( r \) and 4) an identical unitary input waveform (Kuokkanen et al. 2010). If, for example, input waveforms are different, the band-limited SNR generally becomes smaller. The morphological differences between low- and high-BF NL cells (Köppl and Carr 1997; Smith and Rubel 1979) may contribute to the difference in the band-limited SNR, because the existence of dendrites in low-BF cells can alter the summation of synaptic inputs (Agmon-Snir et al. 1998; Grau-Serrat et al. 2003). In gerbil MSO, however, low-voltage-activated potassium conductances counteract the dendritic filtering, resulting in the preservation of the input waveforms along the dendrites (Mathews et al. 2010).

Assuming that the input waveforms are close to identical, the product \( M\lambda_0r^2 \) of the barn owl’s low-frequency SNM should be 3–10 times smaller than the product of the lower bounds used for our estimation (Table 1) to account for the 5- to 10-dB difference in the band-limited SNR (Fig. 4). Since the discharge rates of auditory nerve fibers in response to tones (Köppl and Yates 1999) and the spontaneous spike rates of NM fibers (Köppl 1997b) tend to decrease with increasing BF, it is reasonable to assume that the sound-driven spike rates \( \lambda_0 \) of NM fibers would also decrease with frequency. The frequency dependence of \( \lambda_0 \) may thus not explain the low SNR in the low-frequency NL. Furthermore, even if the spike rate \( \lambda_0 \) is reduced from the lower limit of 300 Hz (Table 1) to 200 Hz, which is the average spontaneous spike rate of NL neurons (Köppl 1997b), at most a factor of 1.5 of the difference in the SNR can be explained. Hence, the low SNRs in low-frequency NL neurons would be more likely to be due to small values of the product \( Mr^2 \). To match theory and experiment, \( Mr^2 \) would need to be at least two to six times smaller than the value predicted from the measured lower bounds of \( M \) and \( r \) in Table 1.

Whereas phase locking of individual NM fibers has already been extensively studied (Köppl 1997a), there are no data available for the population vector strength because simultaneous recordings from multiple NM fibers are required. For this reason, we used the vector strengths of individual NM fibers as an estimate of the population vector strength. If, however, multiple NM fibers are locked to different phases, the population vector strength becomes smaller than that of individual fibers, leading to a decrease in SNR. Furthermore, the lower SNRs in low-frequency NL cells could also be explained.
by a smaller number of independent NM fibers. Thus either the total number of fibers or the actual number of statistically independent fibers may be smaller in low-frequency NL cells than the previous estimate (100–300 fibers/cell; Carr and Boudreau 1993).

Other factors. Increases in stimulus sound intensity generally lead to increases in the spike rate of NM neurons (Peña et al. 1996). Thus the band-limited SNR measured in NL should also increase with sound intensity. This prediction should be tested in future experiments. Our previous modeling results suggested that a high-frequency NL neuron requires substantial sodium conductance in the axon, specifically the first node, to sense sound analog potentials of a few millivolts (Funabiki et al. 2011). High sensitivity to AC signals, however, can also lead to the vulnerability to noise or ITD-independent fluctuations (Ashida et al. 2007). Therefore, a large number of synaptic inputs in mid-to-high-frequency NL neurons may contribute to stabilizing ITD coding by increasing the SNR. By contrast, cycle-to-cycle variability in low-frequency neurons may lead to the smoothing of the ITD tuning curves (Reyes et al. 1996). Considering that the amplitude of the sound analog potential depends on the BF of the cell (Funabiki et al. 2011) and that NL neurons are highly sensitive to noise (Higgs et al. 2006), efficient noise levels should differ between low-BF and mid-to-high-BF cells (Reyes et al. 1996). Analysis of the benefits and disadvantages of synaptic noise in ITD coding is an important subject for future studies.

Fischer et al. (2008) demonstrated that the ITD computation in the owl’s NL can be predicted by the cross-correlation of binaural inputs. Our present and previous (Funabiki et al. 2011) results are generally consistent with cross-correlation (Christianson and Peña 2007; Fischer et al. 2008; Yin et al. 1987). Since the analyses by Fischer et al. (2008) were performed on the output spike rates of the NL neuron in response to broadband noise stimuli, the nonlinearity of the neuron and the integration of multiple frequency channels should also affect the resulting cross-correlation. Our SNR analysis, however, focuses solely on the synaptic inputs, using single-frequency tones. The high SNRs observed in this study should underlie the mechanisms of ITD computation in NL, although further investigation is needed to understand exactly how SNR and cross-correlation are related to each other.

Summary and conclusion. In this report, we examined synaptic inputs in owl’s NL using a SNR analysis. Whereas the band-limited SNRs from mid-to-high-frequency NL neurons agreed well with theoretical estimates, SNRs of low-frequency NL neurons were significantly smaller than the theoretical estimates. One of the possible reasons for these low SNRs is that owl’s low-frequency (≤2 kHz) NL cells could receive significantly fewer inputs from NM fibers than mid-to-high-frequency (>2 kHz) NL cells. Another possibility is that the population vector strength of low-frequency NM fibers could be much smaller than the vector strength of single inputs. Neurons in the gerbil MSO, the mammalian counterpart of the avian NL, were recently reported to receive less than 10 excitatory synaptic inputs (Couchman et al. 2010), in contrast to the owl NL, where neurons in the 5-kHz region are estimated to receive 100–300 inputs per cell (Carr and Boudreau 1993). Most gerbil MSO neurons are tuned to low frequencies (typically, BF < 1,200 Hz; Pecka et al. 2008). Thus similar computational mechanisms could constrain the number of synaptic inputs in both gerbil MSO cells and owl low-frequency NL neurons.

APPENDIX

A note on selecting a bandwidth in calculating the band-limited SNR. As described in MATERIALS AND METHODS, the band-limited SNR is defined as the ratio of the PSD at the stimulus frequency to that at neighboring frequencies (Eq. 1). To evaluate how the SNR depends on the frequency bandwidth used for estimating the noise level (denominator of Eq. 1), we calculated SNRs with bandwidths from 0.4 to 1.5 kHz. The band-limited SNR gradually decreased with increasing bandwidth (Fig. A1A) because the corresponding PSD decreased as a power law with increasing frequency (e.g., Fig. 3). The average change in SNR, however, was within ±1 dB (Fig. A1B). We also confirmed that if the bandwidth is chosen from between 0.6 and 1.5 kHz, our main conclusions did not change. Thus the selection of the bandwidth used for averaging the noise has only minor effects on the calculation of SNR, especially compared with the neuron-to-neuron variability shown in Fig. 4. Note that since the lowest BF of our data set was 800 Hz, its second harmonic frequency of 1,600 Hz did not interfere with the 1-kHz frequency band of 300–1,300 Hz. If one applies the SNR analysis to neurons with even lower BFs, narrower frequency bands should be used to exclude higher harmonics.

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DISCLOSURES

No conflicts of interest, financial or otherwise, are declared by the authors.

AUTHOR CONTRIBUTIONS


REFERENCES


