Acoustic signal characteristic detection by neurons in ventral nucleus of the lateral lemniscus in mice

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Abstract: Under free field conditions, we used single unit extracellular recording to study the detection of acoustic signals by neurons in the ventral nucleus of the lateral lemniscus (VNLL) in Kunming mouse (*Mus musculus*). The results indicate two types of firing patterns in VNLL neurons: onset and sustained. The first spike latency (FSL) of onset neurons was shorter than that of sustained neurons. With increasing sound intensity, the FSL of onset neurons remained stable and that of sustained neurons was shortened, indicating that onset neurons are characterized by precise timing. By comparing the values of Q_{10} and Q_{30} of the frequency tuning curve, no differences between onset and sustained neurons were found, suggesting that firing pattern and frequency tuning are not correlated. Among the three types of rate-intensity function (RIF) found in VNLL neurons, the proportion of monotonic RIF is the largest, followed by saturated RIF, and non-monotonic RIF. The dynamic range (DR) in onset neurons was shorter than in sustained neurons, indicating different capabilities in intensity tuning of different firing patterns and that these differences are correlated with the type of RIF. Our results also show that the best frequency of VNLL neurons was negatively correlated with depth, supporting the view point that the VNLL has frequency topologic organization.

Keywords: Ventral nucleus of the lateral lemniscus; Firing pattern; Frequency tuning; Intensity tuning; Mouse

The ventral nucleus of the lateral lemniscus (VNLL) is the largest nucleus of the lateral lemniscus (LL) (Batra & Fitzpatrick, 2002). As a station in the auditory ascending pathway, the VNLL plays a vital role in acoustic signal processing by receiving afferent projections from the contralateral ventral cochlear nucleus (VCN), ipsilateral trapezoid body (TB), superior olivary complex (SOC) and contralateral dorsal cochlear nucleus (DCN), and casts efferent projections into the inferior colliculus (IC) (Benson & Cant, 2008; Schofield & Cant, 1997; Kelly et al, 2009; Huffman & Covey, 1995; Prather, 2013).

Electrophysiological studies on the VNLL indicate that VNLL neurons can detect, process and encode the characteristics of acoustic signals and have various responding reactions. The firing patterns of VNLL neurons include onset firing (instantaneous firing to the onset of acoustic signals) and sustained firing (sustained firing during the whole interval of acoustic signals) (Zhang & Kelly, 2006). Recently, it was reported that the firing pattern and ability of VNLL neurons to detect acoustic signal characteristics are closely correlated. For example, the first spike latency (FSL) of onset neurons is short and with increasing sound intensity, the FSL shortens insignificantly, whereas those of the sustained neurons are opposite (Zhang & Kelly, 2006). The minimal threshold (MT) of onset neurons is also higher than that of sustained neurons (Aitkin et al, 1970). Regarding firing tuning in VNLL neurons, the value of Q_{10} (Q_{10}=best frequency/the band width at 10 dB above threshold) is used to assess the sharpening of the frequency tuning cures (FTC). Studies on different animals show that the value of Q_{10} in onset neurons is smaller than in sustained neurons (Aitkin et al, 1970; Covey & Casseday, 1991). Regarding intensity tuning in VNLL neurons, the dynamic range (DR, according to the

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rate-intensity function, RIF, of neurons, DR is usually set at 10% higher than the smallest firing spikes and 10% lower than the highest firing spikes) is used to assess intensity tuning abilities. The study on Dutch-belted rabbits (*Oryctolagus cuniculus*) indicates that the DR of onset neurons is smaller than that of sustained neurons (Batra & Fitzpatrick, 1999).

We assume that the firing pattern, intensity and frequency of VNLL neurons have some sort of connection. Despite being an animal model for acoustic studies, detection characteristics and frequency topologic organization of VNLL neurons to acoustic signals in Kunming mice (*Mus musculus*) remain unclear. Here, using single unit extracellular recording, we observed the firing patterns of VNLL neurons and compared FSL, frequency and intensity tuning characteristics under different firing patterns. Our results are helpful in more fully understanding correlations between VNLL neuron firing patterns and acoustic signal detection characteristics, and exploring the physiological functions of VNLL neurons and the effects of ascending projections on the IC.

MATERIALS AND ANIMALS

Animal surgery and electrode preparation

Kunming mice (n=54, 18−28 g) with normal hearing were obtained from the Laboratory Animal Center, Institute of Hubei Preventive Medicine, China. Animal surgical procedures are referenced in previous studies (Wang et al 2007; Tang et al, 2007). All experiment procedures were performed strictly in accordance with the guidelines published in the NIH Guide for the care and use of Laboratory Animals. Anesthesia was induced by injection of mebumal sodium (45−50 mg/kg, bw). After the mouse was immobilized in a stereotaxic apparatus, the VNLL was exposed (4.16−4.48 mm posterior to the bregma, 1.10−1.95 mm lateral to the midline, 2.85−4.10 mm in dorsal ventral depth) by removing skull and dura over the VNLL (Paxinos & Franklin, 2001). An electrode was inserted into the small hole (200−500 μm in diameter) over the VNLL. After surgery, the mouse was transferred onto a shake proof bench in the anechoic shielded chamber and its head was immobilized on a homemade bracket with the aid of a tack (1.8 cm in length) stuck on the skull. The animal's eyes, ears and the center of the speaker were at the same level. Background noise inside the anechoic shielded chamber was 29 dB SPL and the temperature was set at 25−28 ℃. During the experiment, 0.6% mebumal sodium solution was used to keep the animal anesthetized and paralyzed.

By referencing previous studies (Cheng et al, 2013; Mei et al, 2012), a homemade bipolar tungsten electrode made of two insulated tungsten wire electrodes (FHC lnc, Bowdoin, ME, USA) was used to record neuronal responses of the VNLL and to confirm the recording site after experimentation. The electrode was <10 μm in diameter, 1−5 MΩ in impedance and the two poles were 30−80 μm apart.

Acoustic signal system and neuronal response recording

Under free field conditions, the speaker was placed at 0° in the pitching position, and 60° in the contralateral meridional direction (opposite to the animal's external auditory canal opening). Acoustic signals were produced by the Tucker-Davis Technology System 3 (Alachua, USA). Parameters, such as frequency, intensity and duration were controlled by computer and calibrated by a sound meter (2610, B&K, Denmark) and 1/4-inch microphone (4936, B&K, Denmark). Acoustic signals were tone bursts with a duration of 40 ms, rise and fall of 5.0 ms, and a frequency of 2 stimuli/s. The bipolar tungsten electrode was vertically inserted into the VNLL via a hydraulic drive (Kopf 640, USA). Tone burst was adopted to locate acoustic signal sensitive neurons. The responding characteristics of the neurons of the dorsal nucleus of the lateral lemniscus (DNLL) (unpublished data) were used as parallel comparisons to improve accuracy when locating the nucleus. Neuronal responses were amplified by bioelectric amplifier (ISO-DAM, WPI, USA) via the bipolar tungsten electrode, displayed on the oscilloscope (PM3084, FLUKE, USA), monitored by the monitoring apparatus (AM9, GRASS, USA) and then stored in a computer. The depth, best frequency (BF) and MT of the acoustic signal sensitive neuron were recorded. Each tone burst stimulus was repeated 32 times and the induced neuronal firings were profiled into a peri-stimulus time histogram (PSTH). Under the BF, starting from the MT, firing times with the intensity of acoustic signal increasing every 10 dB segment were recorded to infer the RIF. Meanwhile, the highest and lowest frequencies of the neuron were found and their FTCs were inferred.

Electrical burning and histological examination

At the end of the experiment, the recording site in

the anesthetic mouse was burned for 10 s by a 0.1 mA direct current for precise location later. Current stimulation was generated by a stimulator and introduced to the mouse via a direct current stimulation isolator (Model SEN-7203, Nihon Kohden Co, Shinjuku, Tokyo, Japan) and the bipolar tungsten electrode. After a 2 h perfusion with 200 mL saline water and 4% paraformaldehyde, the brain was isolated and fixed in 4% paraformaldehyde for 24 h. Regular paraffin sections (40 μm) were prepared with a vibration slicing machine (VT 1000S, Leica CO, USA) and under Nissl's staining (toluidine blue) for immunohistochemistry examination. The burned location on the section was observed under a light microscope (Leica, DM4000B, Germany) and photographed.

Statistical analysis

All statistical analyses were completed using SPSS 13.0 and all figures were generated with Sigmaplot 10.0. Data are expressed as mean±*SD*. One-way ANOVA and *t*-tests were used to compare differences.

RESULTS

Characteristics of VNLL neurons

For the 98 VNLL neurons we recorded in this study, the dorsal ventral depth was 3 368±156.2 μm (2 844−4 090 μm); BF was 19.89±11.12 kHz (2.6−39 kHz); MT was 52.66±13.11 dB SPL (13.83−71.61 dB SPL); and FSL was 5.78±2.79 ms (2−18.5 ms). Linear regression analysis showed linear relationships between BF and dorsal ventral depth (R=−0.36, P<0.001) (Figure 1A). We found that neurons sensitive to high frequencies are located in the dorsal VNLL. From dorsal to ventral, the BF of VNLL neurons decreased, indicating frequency topological organization along the dorsal-ventral-axis of the VNLL. No linear relationships were found among FSL, MT and dorsal ventral depth (R=0.1, P>0.05; R=0.06, P>0.05) (Figure 1B, C).

Temporal firing pattern of VNLL neurons

As one of the basic features of central auditory neurons, the temporal firing pattern reflects firing changes in neurons before and after stimulation. With acoustic signals at BF and MT+20 dB, the firing patterns of 98 VNLL neurons were recorded. Following the research of Zhang & Kelly (2006), our observed firing patterns were categorized into two types: onset and sustained. The onset type can be sub-categorized into: (1) phasic: only one or two spikes to the onset of the acoustic signals; (2) phasic burst: several spikes to the onset of the acoustic signals; and (3) double burst: fixed intervals can be found between the spikes to the onset of the acoustic signal. Likewise, the sustained type can also be sub-categorized into: (1) onset plus sustained: significant response can be found to the onset of the acoustic signals,

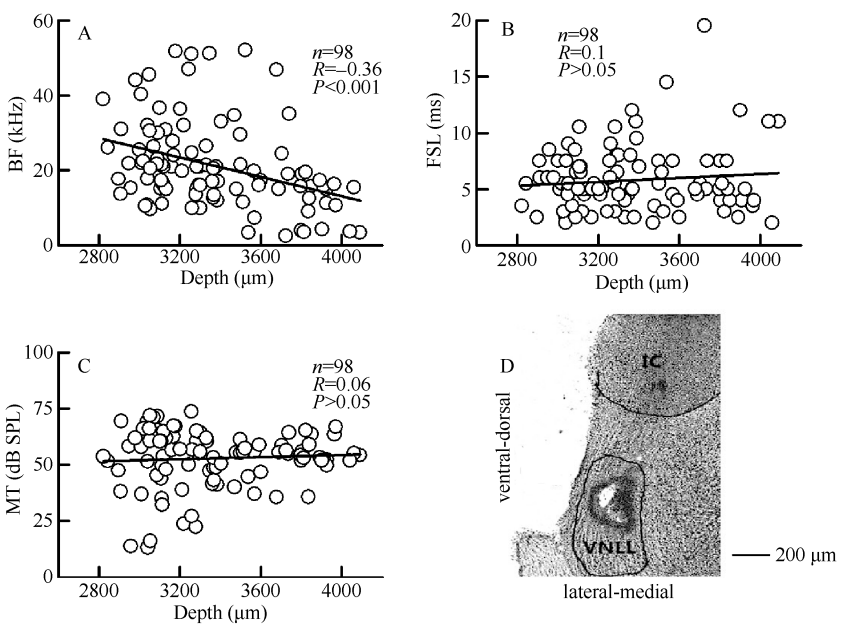

Figure 1 Correlations between recording depth and BF (A), FSL (B) and MT (C) in VNLL neurons and Nissl-stained coronal section of the VNLL and surrounding structures (D)

n: Numbers of neurons; R: correlation coefficient.

then decreases and lasts through the entire duration; (2) tonic: a fixed firing rate lasts through the entire duration; and (3) primary-like: the firing rate to the onset of the acoustic signals quickly reaches maximum, then decreases to a certain level. The representative post-stimulus time histograms (PSTH) of these six firing patterns and neuron numbers are shown in Figure 2. Among them, phasic burst had the highest percentage, followed by tonic, phasic and onset plus sustained, respectively; primary-like and double burst had the smallest percentages. Although differences were found among the percentages of various firing patterns, the quantities of onset (52/98, 53.06%) and sustained (46/98, 46.94%) patterns were comparable.

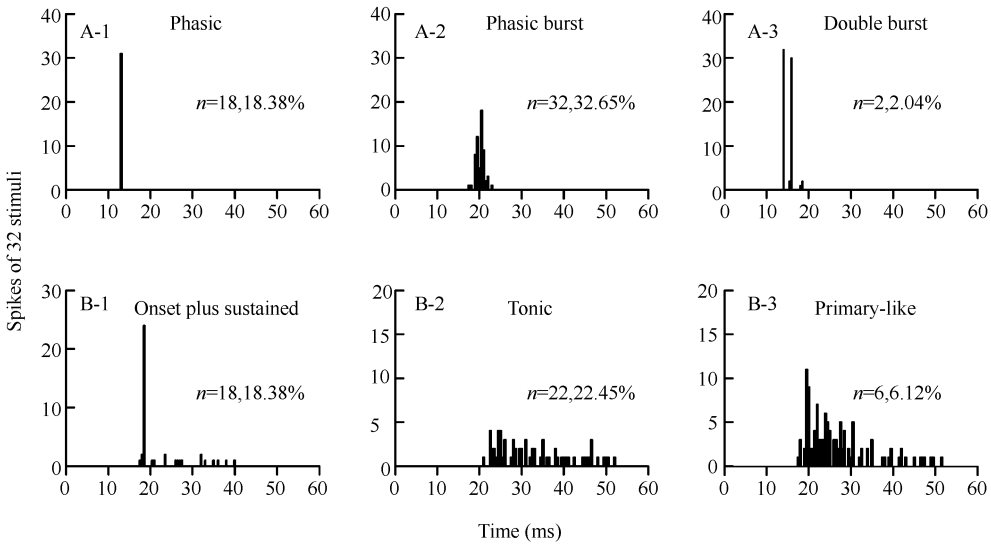

Figure 2 Types of onset (A-1, A-2, A-3) and sustained (B-1, B-2, B-3) patterns of recorded VNLL neurons

n: Numbers of neurons; Recording depth (μm), MT (dB SPL) and BF (kHz) of these neuron are 3118, 53.83, 16.2 (A-1); 3739, 64.28, 35 (A-2); 3970, 66.79, 10.6 (A-3); 2908, 38.14, 13.7 (B-1); 4090, 54.22, 3.4 (B-2); 3042, 3.14, 17.8 (B-3), respectively.

Under intensity at MT+20 dB, the FSL of onset VNLL neurons was 5.47±1.86 ms (2−11 ms), with 67.31% of the FSL within the range of 2.5−5 ms; the FSL of sustained VNLL neurons was 7.1±3.13 ms (2−19.5 ms) with a relatively wide distribution (Figure 3A, C). The FSL of onset VNLL neurons was shorter than that of sustained neurons (P<0.001, unpaired t-test). When the intensity of the acoustic signal increased every 10 dB segment to MT+50 dB, no significant changes in the FSL of onset VNLL neurons were observed (one-way ANOVA, P>0.05), whereas, the FSL of sustained neurons decreased (one-way ANOVA, P<0.001) (Figure 3B, D).

Frequency tuning of VNLL neurons

Frequency is a basic parameter of acoustic signals. Auditory neurons analyze and encode external acoustic stimulation, which play an important role in the behavior of humans and other animals. FTC are commonly used in assessing the frequency tuning of auditory neurons. According to Suga (1997) and our previous data, the FTC of the 71 VNLL neurons recorded here were divided into six types: (1) V-shape: with increasing intensity, the verges of high and low frequency are broadening; (2) U-shape: the changes in FTC are independent of intensities [the V- and U-shape can be differentiated by bandwidth level intolerance (BLI, $BLI=BW\text{max}/BW10$), if BLI<3, it is U-shape, if $BLI\geqslant$3, it is V-shape]; (3) Lower-tail-upper-sharp (LTUS): the low frequency verge is characterized with a long tail and the high frequency verge is very sharp; (4) Upper-tail-lower-sharp (UTLS): the high frequency verge is characterized with a long tail and the low frequency verge is very sharp [in LTUS and UTLS, the $\text{Slope}_{MT+30-MT+10}\geq$30%, otherwise, if $\text{Slope}_{MT+30-MT+10}$<30%, the FTC is V- or U-shape]; (5) W-shape: two peaks can be found in FTC and the two frequency tuning segments are partitioned by an auditory completely insensitive area only under low intensity, but not under high intensity; and (6) Double V-shape: two peaks can be found in FTC and the two frequency tuning segments are partitioned by an auditory completely insensitive area under both low and high intensities (Figure 4).

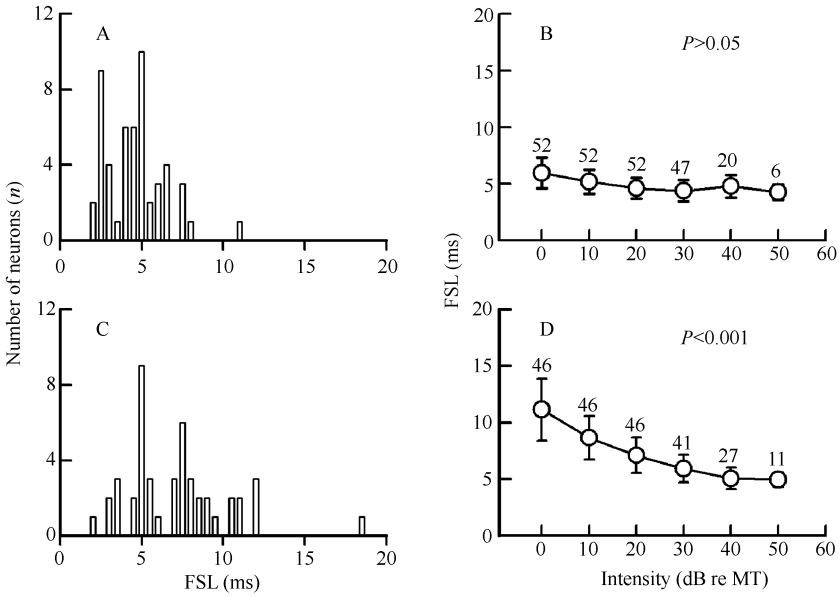

Figure 3 Distribution of FSL in onset neurons (A) and sustained neurons (C) and FSL-intensity function in onset neurons (B) and sustained neurons (D)

Abscissa of B and D were normalized by each neuron's MT as the control of tone intensity; numbers above the vertical bars in B and D are the numbers of neurons calculated statistically.

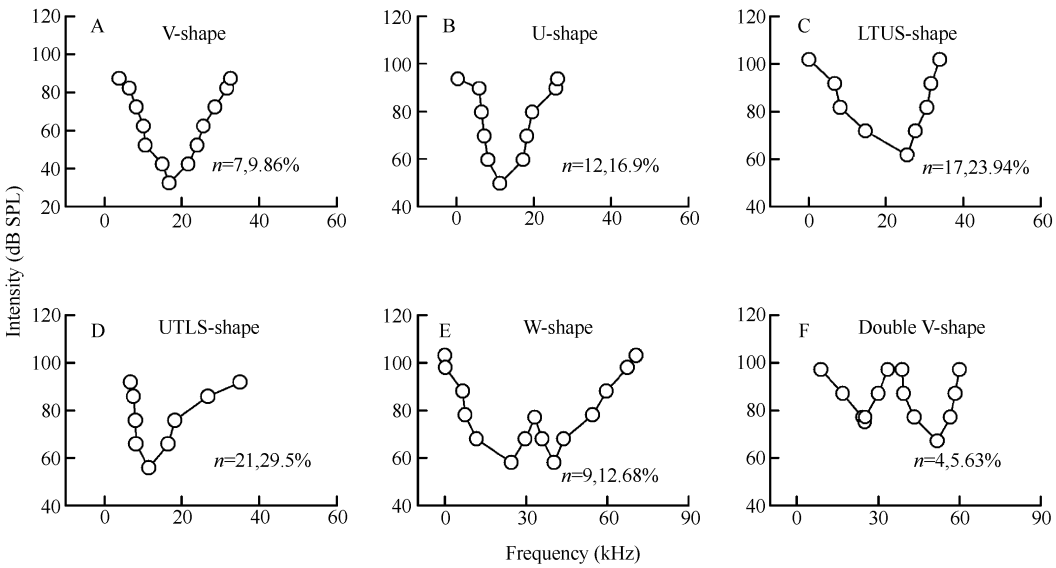

Figure 4 Types of frequency tuning curves of recorded VNLL neurons

n: Numbers of neurons; Recording depth (μm), MT (dB SPL) and BF (kHz) of these neuron are 3114, 32.22, 16.8 (A); 3925, 49.63, 11.2 (B); 3001, 61.66, 25.5 (C); 3513, 55.81, 11.5 (D); 3009, 58.03, 40.3 (E); 3179, 67.09, 51.8 (F), respectively.

Among the 71 neurons, 38 were onset and 33 were sustained. The MT of the onset neurons was 55.33±10.83 dB SPL (27.09−69.34 dB SPL) and that of the sustained neurons was 48.83±12.85 dB SPL (13.83−73.54 dB SPL) (P<0.05, unpaired t-test) (Figure 5 A). The value of Q_{10} of the FTC of onset neurons was 2.49±1.67 (0.65−7.01) and that of the sustained neurons was 3.43±3.83 (0.8−19.26) (P>0.05, unpaired t-test), whereas, the value of Q_{30} of the FTC of onset neurons was 0.94±0.52 (0.26−2.45) and that of the sustained neurons was 0.96±0.26 (0.13−1.99) (P>0.05, unpaired t-test) (Figure 5 B, C), indicating that the firing patterns and frequency tuning of VNLL neurons are irrelevant.

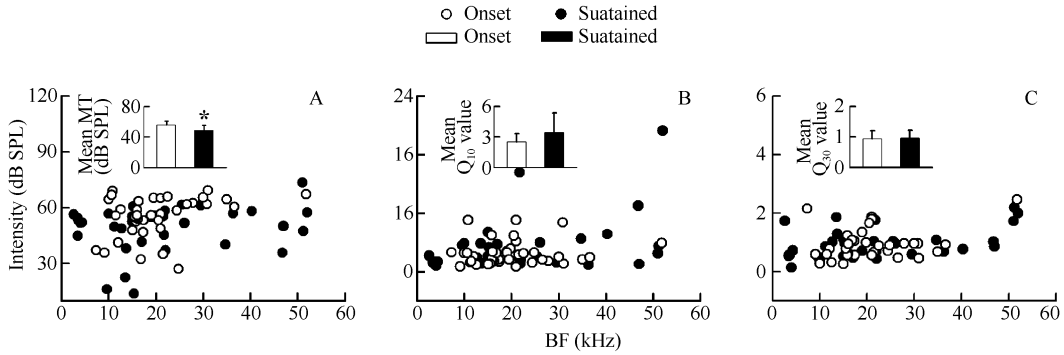

Figure 5 Differences in the MT (A), Q_{10} value (B) and Q_{30} value (C) between onset neurons and sustained neurons

*: $P<0.05$.

Intensity tuning of VNLL neurons

Intensity reflects the amplitude of acoustic vibration. Within certain range, most of the auditory neurons increase spiking with increasing intensity. Under relatively high intensity, different neurons show different changes in firing rate function. With an acoustic signal at BF, 40 ms, and intensity increases every 10 dB segment, the firing numbers of VNLL neurons changed and showed various characteristics of intensity tuning. According to previous work and our own data (Tang et al, 2007; Zhou & Jen, 2000), under increasing intensity, if the firing numbers keep increasing, neurons are recognized as monotonic RIF; if the firing numbers do not change or decrease to <25%, neurons are recognized as saturated RIF; and if the firing numbers decrease to≥25%, neurons are recognized as non-monotonic RIF. The representative curves of these three types of RIF and numbers of neurons are shown in Figure 6. The monotonic RIF had the highest percentages, followed by saturated and then non-monotonic RIF.

Of the 91 VNLL neurons we recorded in this study, 49 were onset and 42 were sustained. The percentages of monotonic, saturated and non-monotonic RIF of onset neurons were 48.98% (24/49), 42.86% (21/49) and 8.16% (4/49), respectively; of sustained neurons the percentages were 61.9% (26/42), 21.43% (9/42) and 16.67% (7/42), respectively (Figure 7A). Therefore, in onset and sustained neurons, the distribution patterns of RIF from high to low were monotonic, saturated and non-monotonic, respectively. The DR of the onset neurons was smaller than that of sustained neurons ($P<0.01$, unpaired *t*-test). In neurons with monotonic and saturated RIF, the DR of the onset neurons was smaller than sustained neurons ($P<0.01$; $P<0.05$, unpaired *t*-test); in neurons with non-monotonic RIF, no differences were found between onset and sustained neurons ($P>0.05$, unpaired *t*-test) (Figure 7B).

DISCUSSION

Firing patterns and temporal features of VNLL neurons

The percentages of onset and sustained patterns were comparable, 53.06% (52/98) and 46.94% (46/98), respectively, consistent with previous findings on big brown bats (*Eptesicus serotinus*) (Covey & Casseday, 1991), rats (Zhang & Kelly, 2006) and rabbits (Batral & Fitzpatrick, 1999). The 2.04% (2/98) double-burst pattern we found has only been reported in rats (Zhang &

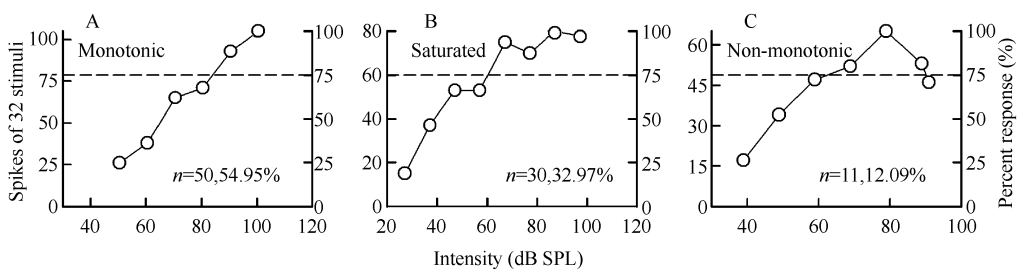

Figure 6 Types of rate-intensity function of recorded VNLL neurons

n: Numbers of neurons; Recording depth (μm), MT (dB SPL) and BF (kHz) of these neuron are 3403, 50.52, 33 (A); 3259, 27.09, 24.8 (B); 3211, 38.89, 19.8 (C), respectively.

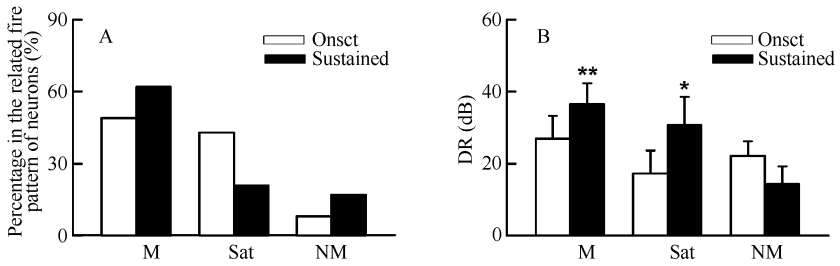

Figure 7 Differences in percentage of RIF (A) and DR (B) between onset neurons and sustained neurons

M: monotonic; Sat: saturated; NM: non-monotonic. *: $P<0.05$; **: $P<0.01$.

Kelly, 2006) and its biological meaning in acoustic signal processing remains unclear.

The FSL of onset neurons was shorter than sustained neurons. With the increase in intensity, the FSL of onset neurons remained stable (Figure 3B), indicating a feature of precise timing of onset neurons. This specific feature of onset VNLL neurons may be attributable to projections of octopus cells in the cochlear nucleus (CN) (Adams et al, 1997; Pollak et al, 2011). The octopus cell have large, irregularly shaped cell bodies with three to five thick primary dendrites emerging from only one side of the cell body. The sizable low voltage activated potassium channels on its membranes endow the cell with a low impedance and fast time constant, which bestow its projections to the VNLL with temporal advantages (Johnston et al, 2010; Bal & Ocrtel, 2007). The precise timing of onset VNLL neurons is not only beneficial in the encoding of acoustic signal onset, but also affects the process of IC neurons to temporal information via its ascending projections. For example, studies on rats (Nayagam et al, 2005), Mexican free-tailed bats (*Tadarida brasilensis mexicana*) (Xie et al, 2007) and big brown bats (Voytenko & Galazyuk, 2008) show that an inhibitory postsynaptic potential (IPSP) can be observed ahead of the excitations of some IC neurons, which means an IPSP occurs before depolarization. It is assumed that because of the temporal advantage of VNLL neurons possess, the inhibitory projection from VNLL neurons will reach the IC ahead of the excitatory projection, thereafter, an early IPSP is initiated.

The FSL of sustained VNLL neurons decreased with increasing intensity (Figure 3D). This phenomenon is not specific to sustained VNLL neurons (Zhang & Kelly, 2006) and has also been observed in the CN (Goldberg & Brownell, 1973), SOC (Sanes & Rubel, 1988), IC (Tan et al, 2008) and auditory cortical (AC) (Heil, 1997). Therefore, it is assumed that FSL encodes the intensity of the onset of the acoustic signal. The larger the intensity, the shorter the FSL, and the quicker the signal can be recognized. However, correlations between FSL and intensity are unclear. Pharmacological studies show that neither ɣ-aminobutyric acid (GABA) nor glycine (Gly) affects correlations between FSL and intensity in AC and IC neurons (Zhang et al, 2005), indicating that these correlations may be encoded by auditory neurons below the IC, but this requires further exploration.

Frequency tuning and frequency topologic organization of VNLL neurons

Although the percentages of six different types of FTC are consistent with previous studies on rats (Zhang & Kelly, 2006) and rabbits (Batra & Fitzpatrick, 1999), the degrees of sharpening are quite different among different species. For example, in this study, the average value of Q_{10} of the FTC of VNLL neurons in Kunming mice was 2.9, but was 7.1 (Zhang & Kelly, 2006) in rats and 9.1 (Covey & Casseday, 1991) in bats, respectively, indicating that different species are characterized with different capabilities of frequency tuning. The sharpening of FTC and GABAergic inhibitory projection are correlated with the formation of the inhibitory area of frequency tuning (Chen & Jen, 2000). The FTC of VNLL neurons in mice have smaller Q_{10} and low degrees of sharpening. Because the VNLL in mice receives less inhibitory projections from the ipsilateral trapezoid body, but has a wide frequency receptive range to excitatory projections from the contralateral cochlear nucleus, perhaps the FTC of VNLL neurons is not characterized by significant sharpening.

No differences were found in the Q_{10} of the FTC in onset and sustained neurons (Figure 5). However, studies on big brown bats and cats show that the Q_{10} in onset neurons is smaller than sustained neurons (Covey & Casseday, 1991; Aitkin et al, 1970). Based on these discrepancies, it is assumed that different species have different neural circuits corresponding to specific behaviors.

For example, because the frequency tuning of the onset VNLL neurons in Mexican free-tailed bats affects the reorganization of IC neurons to some special communication signals, they show certain corresponding firing patterns and features of frequency tuning (Pollak et al, 2011).

Frequency topological organization of VNLL neurons remains controversial. When the IC of rats was infected with retrograde tracing agents, low frequency neurons in the VNLL distributed along the band, whereas, high frequency neurons were evenly distributed (Kelly et al, 1998). Electrophysiological experiments on bats (Covey & Casseday, 1991) and cats (Aitkin et al, 1970) show that low frequency neurons are distributed dorsally of the VNLL, whereas, high frequency neurons are distributed ventrally of the VNLL. Although differences exist in the distribution patterns of different species, the logical distribution of low and high frequency neurons supports the view of frequency topologic organization in the VNLL. However, an electrophysiological experiment on rats (Nayagam et al, 2006) and the study on infecting retrograde tracing agents in the IC of cats (Glendenning & Hutson, 1998) and the primary auditory field (AI) of Mongolian gerbils (Budinger et al, 2013) indicate a lack of topologic organization in the VNLL. Possible reasons of these discrepancies are differences in animal models and experimental methods. In this study, the BF and dorsal ventral depth of VNLL neurons were negatively correlated (Figure 1A). Neurons with high frequencies were located dorsally of the VNLL and from dorsal to ventral, the BF of neurons decreased, indicating frequency topologic organization along the dorsal-ventral-axis of the VNLL.

Intensity tuning of VNLL neurons

In the 91 VNLL neurons recorded here, the percentage of non-monotonic RIF (12.09%) was lower than that in the advanced auditory nuclei of mice, such as the IC (Tang et al, 2007) and AC (Qi et al, 2013). The occurrence of non-monotonic type is due to inhibitory input tuning (Chen & Jen, 2000; Andrew & Ellen, 2009 Wu et al, 2006; Tang et al, 2008). The low level of non-monotonic RIF in VNLL neurons may be consistent with less inhibitory inputs in the VNLL. Previous histochemical studies show that excitatory projections in the VNLL are mainly from the CN (Suneja et al, 1995), whereas low levels of inhibitory projections are mainly from the TB (Yavuzoglu et al, 2010).

The intensity tuning of onset and sustained neurons were compared and values of DR were used as an index to assess the sensitivity of neurons to acoustic signal intensity. The smaller the DR, the narrower the intensity tuning and the higher the sensitivity, meaning that variation in intensity can induce significant changes in the neuronal spiking of humans and other animals. The smaller DR in onset neurons compared to sustained neurons indicates different sensitivity in these two types of neurons and implies that the intensity encoded by neuronal impulses is affected by neuronal firing patterns. Although previous studies have reported similar phenomena in the VNLL (Batra & Fitzpatrick, 1999), DNLL (unpublished data) and IC (Bal et al, 2002), if differences in DR can be observed among neurons with different firing patterns then correlations with RIF remain unclear. The DR of two neuron types with three different RIF was compared. In monotonic and saturated RIF, the DR of onset neurons was smaller than sustained neurons, but in non-monotonic RIF, no differences were found (Figure 7B), indicating that DR is correlated with RIF type and in general, onset neurons are more sensitive than sustained neurons in intensity tuning.

The firing patterns of VNLL neurons are affected by multiple factors and truthfully reflect their ability in intensity tuning, The firing patterns of certain VNLL neurons change with changes in acoustic signals (Zhang & Kelly, 2006) indicating that their firing pattern and intensity are regulated by different signal pathways and integrations of excitatory and inhibitory inputs within the time window.

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