The severity of developmental hearing loss does not determine the magnitude of synapse dysfunction

TODD M. MOWERY, VIBHAKAR C. KOTAK, AND DAN H. SANES*

Center for Neural Science, New York University, New York, New York, USA

The loss of auditory experience can disrupt synapse function, particularly when it occurs during development. However, the extent of hearing loss can vary from mild to profound, and the duration of hearing loss can vary from days to years. Here, we asked whether the dysfunction of central auditory synapses scales with the severity of hearing loss. The manipulations range from mild sound attenuation to complete deafferentation at the time of hearing onset. Synapse function is measured in central auditory structures from the cochlear nucleus to cortex. The core finding is that even a ~25 dB attenuation in sensation level produces a quantitatively similar change to synaptic currents and membrane properties, as compared to deafferentation. Therefore, profound changes to central processing may occur even when developmental hearing loss is mild, provided it occurs when sound is first transduced.

INTRODUCTION

Many functional properties of central auditory neurons are use-dependent. They can be altered by passive exposure to sound, as well as active experiences such as learning. Central auditory plasticity is particularly evident throughout development during which it supports normal maturation of auditory processing (Sanes and Bao, 2009; Sanes and Woolley, 2011). However, this sensitivity to auditory experience can also introduce a risk: when sound-evoked activity is reduced due to the loss of hearing, both synaptic and membrane properties can assume a dysfunctional state (for a recent review, see Sanes, 2013). Much of primary evidence in support of this theory emerges from experiments in which the cochlea is damaged or removed. For example, our work on bilateral hearing loss has examined the synaptic consequences in the superior olive, inferior colliculus, and auditory cortex (e.g., Kotak and Sanes, 1997; Vale and Sanes, 2000; Vale and Sanes, 2002; Vale *et al.*, 2003; Kotak *et al.*, 2005). Therefore, the magnitude of functional changes in the auditory circuits during less severe forms of hearing impairment such as during middle ear damage has not been thoroughly explored.

In this review, we explore the issue of hearing loss severity by comparing cellular measures obtained following different experimental manipulations to the auditory periphery. For developmental hearing loss, these studies suggest that mild hearing loss can produce changes to cellular properties that are similar to those observed following cochlear damage. These results suggest that auditory deprivation during a

*Corresponding author: sanes@cns.nyu.edu

Proceedings of ISAAR 2013: Auditory Plasticity – Listening with the Brain. 4th symposium on Auditory and Audiological Research. August 2013, Nyborg, Denmark. Edited by T. Dau, S. Santurette, J. C. Dalsgaard, L. Tranebjærg, T. Andersen, and T. Poulsen. ISBN: 978-87-990013-4-7. The Danavox Jubilee Foundation, 2014.

sensitive time window at hearing onset (postnatal day 11 in gerbil) leads to functional deficits in the brain independent of the severity of peripheral dysfunction. Further, they imply that behavioural delays reported for transient hearing loss in humans could be attributable to central nervous system alterations.

NORMAL DEVELOPMENT OF AUDITORY SYNAPSE FUNCTION

The functional properties of synapses have been measured as a function of age in several species. The measurements are commonly made from slice of tissue through a central auditory structure that is maintained in a warm, oxygenated saline solution for several hours. The synaptic responses are obtained with whole-cell current- or voltage clamp recordings, which allow one to make direct measurements of synaptic potentials or currents, respectively. Both evoked and spontaneous synaptic events can be quantified, with the most common parameters being amplitude and kinetics.

Both central excitatory and inhibitory synapses are functional well before the onset of hearing. In rodents, evoked synaptic responses can be observed in tissue obtained at birth, whereas sound transduction by the cochlea is first observed over one week later (Sanes and Walsh, 1997; Fitzgerald and Sanes, 2001). In fact, spontaneous action potentials are also observed in the auditory central nervous system (CNS) before sound first activates the cochlea. These action potentials may be evoked by hair cell activity (Jones *et al.*, 2007; Tritsch *et al.*, 2007; Johnson *et al.*, 2011), and by mechanisms that are intrinsic to the CNS (Kotak *et al.*, 2007b; Tritsch *et al.*, 2010; Kotak *et al.*, 2012). In either case, there is a great deal of spontaneous synaptic transmission in the auditory CNS prior to the onset of sound-evoked responses.

The amplitude and decay time of synaptic potentials mature rapidly beginning at about the time of ear canal opening in rodents (about 9-12 days postnatal, depending on the species). Figure 1 illustrates the developmental time course for excitatory and inhibitory postsynaptic potentials (EPSPs and IPSPs) recorded in the mouse auditory cortex (Oswald and Reyes, 2008; 2011). The measurements of amplitudes and decay times demonstrate that maturation continues after the onset of hearing and an adult-like state is reached within a few weeks. Many of the observed functional changes are highly significant, suggesting that mature auditory processing depends on sufficient development of synapse function.

Recordings obtained in auditory brainstem nuclei are generally consistent with this rate of development. Within about two weeks of hearing onset, EPSPs and IPSPs recorded in the lateral and medial superior olivary nuclei display adult-like kinetics (Sanes, 1993; Kandler and Friauf, 1995; Scott *et al.*, 2005; Magnusson *et al.*, 2005; Chirila *et al.*, 2007). Furthermore, these findings are consistent with observations from many other auditory brainstem nuclei (Chuhma and Ohmori, 1998; Taschenberger and von Gersdorff, 2000; Brenowitz and Trussell, 2001; Balakrishnan *et al.*, 2003; Nakamura and Takahashi, 2007; Gao and Lu, 2008; Sanchez *et al.*, 2010). Although exceptions to this pattern of development are seldom observed, IPSC decay time in cortical pyramidal neurons displays a relatively prolonged maturation, only reaching an adult value at about 3 postnatal

months (Takesian *et al.*, 2012). A late maturation of synaptic inhibition is consistent with the prolonged transition of $GABA_A$ receptor subunit expression in human cortex (Pinto *et al.*, 2010).

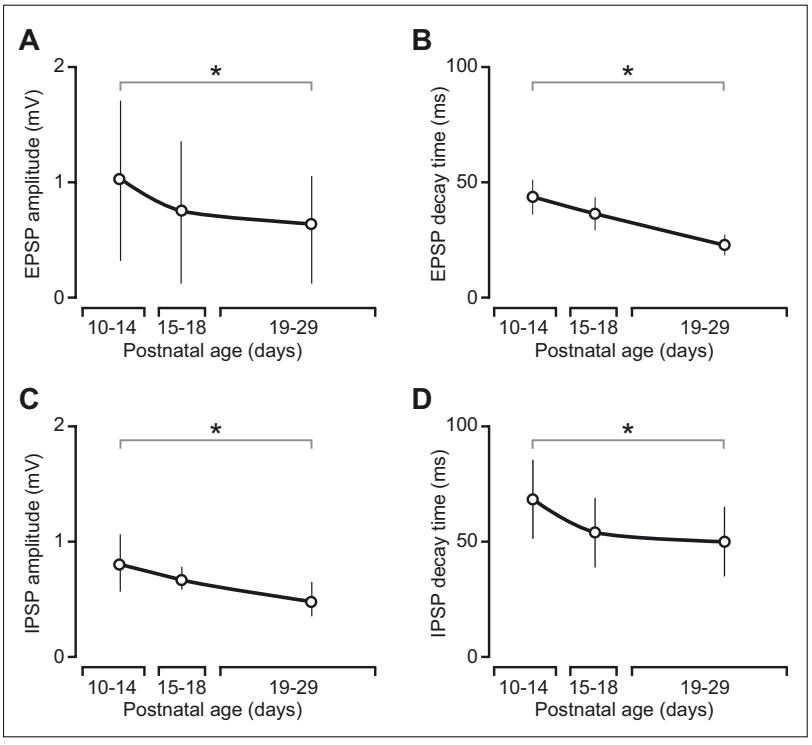

Fig. 1: Excitatory and inhibitory synaptic potential amplitudes and decay times measured from in vitro whole-cell recordings in mouse auditory cortex. Synaptic potentials are in response to stimulation of a single presynaptic neuron. In each graph, mature responses appear to emerge over about 14 days. There is a developmental decrease in (A) EPSP amplitude, (B) EPSP decay time, (C) IPSP amplitude, and (D) IPSP decay time. Asterisks indicate a statistically significant change. Adapted from Oswald and Reyes (2008; 2011).

The wealth of data obtained from recordings in brain slices is consistent with the few in vivo whole-cell studies that have been conducted on anesthetized animals. As shown in Fig. 2A, the decay times for EPSCs and IPSCs are relatively stable after about 25 days postnatal. Similarly, measures of sound-evoked plasticity mature at

about the same rate. In animals younger than P21, sound stimulation leads to an increase in both EPSC and IPSC conductance (Fig. 2B), but this form of plasticity is absent after P25 (Dorrn *et al.*, 2010).

The mechanistic bases for changes in the amplitude of a synaptic response are manifold. For example, the number of neurotransmitter receptors at the synapse, the conductance of single receptor-coupled channels, the amount of transmitter released, and the distribution of ions across the membrane, can each determine the response amplitude. Furthermore, the specific molecular composition of a receptor will establish its mean open time when bound by neurotransmitter, and this will determine the decay time for a synaptic event. Therefore, measurement of synaptic amplitude and kinetics are a logical first step in determining the molecular and genetic basis for the effects associated with hearing loss.

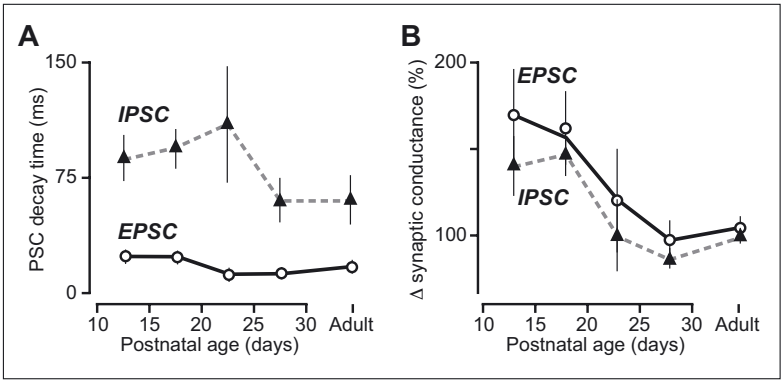

Fig. 2: Synaptic current decay times and sound-induced plasticity measured with in vivo whole-cell recordings from the rat auditory cortex. Mature responses are observed by postnatal day 25-30. (A) The decay times of sound-evoked EPSCs and IPSC decline after P20. (B) An increase in sound-evoked EPSC or IPSC conductance occurs following 3-5 min of sound stimulation, but this phenomenon fails to occur after P25. Adapted from Dorrn *et al.*, (2010).

SOUND ATTENTUATION VERSUS DEAFFERENTATION

Since synaptic responses display a well-characterized maturation in the rodent central auditory system, it is possible to study whether hearing loss induces an impairment. More importantly, it permits for the quantitative comparison of different forms of hearing loss. Conductive hearing loss (CHL), such as that which may occur during bouts of otitis media with effusion, leads to sound attenuation and a smaller neural response to a given SPL. Depending on its severity (e.g., ear canal

atresia vs otosclerosis), the magnitude of sound attenuation can range from 10-50 dB. However, CHL is not associated with a direct injury to the cochlea, and the innervation density should not be altered. In contrast, sensorineural hearing loss (SNHL) involves a direct injury to the cochlea, and would include both a smaller neural response to a given SPL, as well as deafferentation of the CNS.

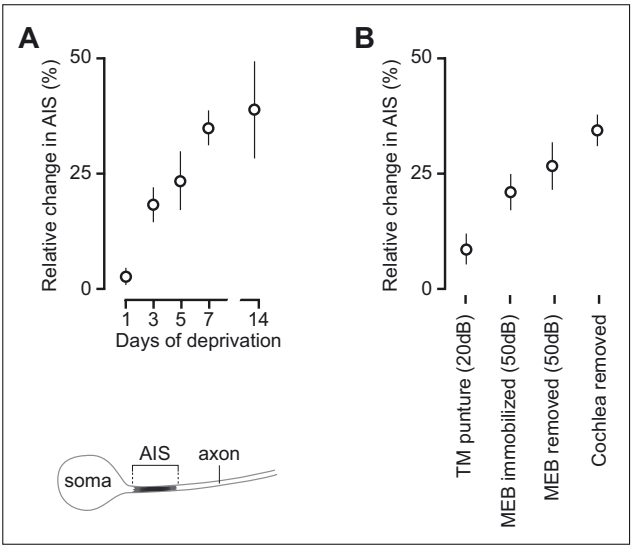

Fig. 3: Effect of hearing loss is correlated with severity in the chick cochlear nucleus. (A) Action potentials are initiated in a small region of membrane containing a high density of voltage-gated sodium channels, called the axon initial segment (AIS). The AIS became more extended along the axon following hearing loss, and the effect size was correlated with the severity of the manipulation. The SNHL-induced alteration of AIS length emerges over about 7 days. (B) The magnitude of AIS expansion is correlated with the severity of hearing loss. The first 3 conditions represent CHL (amount of attenuation shown in parentheses), and the final manipulation represents SNHL. (TM, tympanic membrane; MEB, middle ear bone). All animals were reared in the same acoustic environment. Adapted from Kuba *et al.* (2010).

There is evidence that hearing-loss-induced changes to cellular properties are correlated with the severity of deprivation. In the chick cochlear nucleus, developmental hearing loss causes a redistribution of sodium channels on the axon initial segment (AIS), a region of membrane responsible for action potential initiation. This effect begins to emerge after 1 day of hearing loss, and requires

about 7 days to reach an asymptotic level (Fig. 3A). Furthermore, the magnitude of this change is correlated with the type of experimentally-induced hearing loss (Kuba *et al.*, 2010). As shown in Fig. 3B, there is only a modest change in AIS length in response to a mild CHL (puncture of the tympanic membrane) which induces approximately 20 dB of attenuation, as measured with auditory brainstem response (ABR). However, moderate CHL (middle-ear bone immobilization or removal; ~50 dB of attenuation) leads to a highly significant increase in AIS length. Finally, SNHL (cochlea removal) induces the largest effect. These results indicate that CHL can elicit significant changes to the CNS cellular function, but that the effects due to SNHL are quantitatively larger.

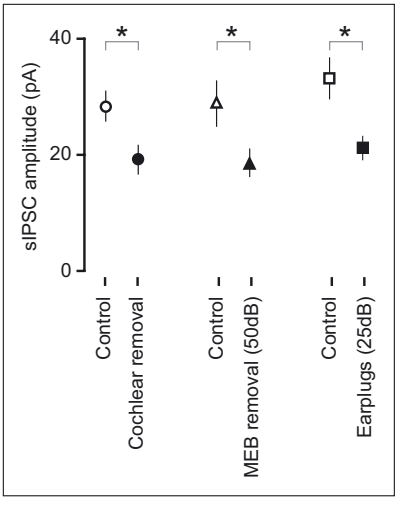

Fig. 4: A comparison of the impact of 3 forms of developmental hearing loss on auditory cortex inhibition. Bilateral cochlear removal, middle ear bone removal, or earplug insertion were induced at postnatal day 10-11, and spontaneous IPSCs were subsequently recorded from auditory cortex pyramidal neurons. IPSC amplitude declined to an equivalent degree in each form of hearing loss, as compared to age-matched control recordings (MEB, middle ear bone). Asterisks indicate a statistically significant change. Adapted from Kotak *et al.* (2008), Takesian *et al.* (2012), and Mowery *et al.* (2013).

Although no single study provides a similar comparison of hearing loss severity as it relates to synaptic function, we have performed identical measures of cortical inhibitory currents following 3 different forms of developmental deprivation (Kotak *et al.*, 2008; Takesian *et al.*, 2012; Mowery *et al.*, 2013). Figure 4 shows the mean

amplitude of spontaneous IPSCs recorded in gerbil auditory cortex from control animals, in comparison to animals reared with SNHL (i.e., bilateral cochlea removal), moderate CHL (i.e, bilateral malleus removal), or mild CHL (i.e., bilateral earplugs). Each form of hearing loss induces a nearly identical decrease in IPSC amplitude. Furthermore, when CHL is induced in adult animals, it does not lead to a decrease in IPSC amplitude. The relative impact of developmental moderate CHL or SNHL on transmitter release has also been examined for both excitatory and inhibitory synapses in auditory cortex (Xu *et al.*, 2007; Takesian *et al.*, 2010). Again, there was little difference between bilateral CHL versus bilateral SNHL. Following either manipulation there was significantly greater synaptic depression in response to multiple stimuli. For excitatory synapses, the SNHL elicited effect was slightly larger than that observed with CHL (Xu *et al.*, 2007). Therefore, cortical synaptic function can be as sensitive to sound attenuation as it is to complete deafferentation.

Although very different manipulations can result in similar outcome measures, there are cases in which the SNHL elicits significantly larger effects. For example, SNHL leads to a greater reduction in spike frequency adaptation in response to trains of injected current pulses, as compared to CHL (Xu *et al.*, 2007). Finally, hearing loss can disrupt long-term synaptic plasticity (Kotak *et al.*, 2007a), a neuronal mechanism thought to be involved in learning. For example, inhibitory synapses in auditory cortex display long-term potentiation following trains of afferent stimulation, and this synaptic plasticity is diminished by hearing loss (Xu *et al.*, 2010). Furthermore, the reduction of plasticity is greater for SNHL than it is for CHL (Fig. 5). These cortical studies suggest that deafferentation can have a greater influence on the development of cellular properties, as compared to manipulations that result in sound attenuation.

SHORT- VERSUS LONG-TERM HEARING LOSS

Although developmental hearing loss has been shown to influence many neural properties, most of these results are obtained following a short survival time. These data demonstrate that the effects can appear within hours to days, but they do not address whether the changes are permanent. Our studies of the impact of hearing loss on synaptic inhibition in auditory cortex suggest that cellular deficits can persist into adulthood. Figure 6 plots the mean amplitudes of spontaneous IPSC recorded from auditory cortex neurons following developmental CHL, as a function of survival time. CHL causes a significant reduction in IPSC amplitude, and this effect is present at both short and long survival times (Takesian *et al.*, 2012). The long duration inhibitory currents that are observed after SNHL resemble IPSCs that are recorded in neurons from pre-hearing animals, suggesting that normal acoustic experience is essential for maturational progress of $GABA_A$ receptor subunit function to occur (Kotak *et al.*, 2008). Specifically, the agonists of $GABA_A$ receptor subunits $\alpha 1$ and $\beta 2/3$ did not produce effects on IPSC kinetics, and this lack of an effect resembled that observed in neurons from pre-hearing animals.

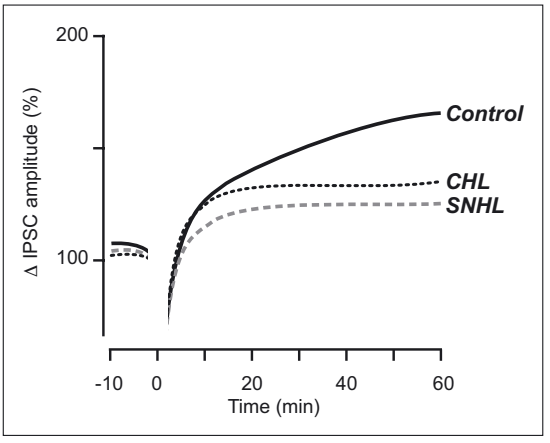

Fig. 5: The impact of hearing loss on inhibitory synaptic long-term potentiation recorded in the gerbil auditory cortex. Evoked IPSCs were recorded from pyramidal neurons for 10 minutes, followed by a series of stimulus trains that were designed to emulate the temporal discharge pattern of auditory cortex neurons in vivo. Following this treatment, the amplitude of evoked IPSCs was potentiated by 155%, as compared to the pre-treatment value (Control). The magnitude of this potentiation was much smaller for animals with developmental hearing loss. However, SNHL resulted in a greater effect, as compared to CHL. Adapted from Xu *et al.* (2010).

One mechanistic explanation for this effect is that adult $GABA_A$ receptor subunits are not properly trafficked to the synaptic membrane (Sarro *et al.*, 2008). Since CHL also causes IPSC decay times to remain longer, both at short and long survival times, the functional expression of $GABA_A$ receptors may remain compromised for the duration of hearing loss. In this regard, it is interesting to note that inhibitory maturation can be induced with pharmacological manipulations that boost GABAergic transmission, such that normal IPSC amplitudes and kinetics are observed in animals that remain deafened (Kotak *et al.*, 2013).

Although impairment of cellular function can persist during ongoing hearing loss, many cellular properties are normal after long survival times. Recordings obtained from adult cortical inhibitory interneurons following developmental CHL indicate that passive membrane properties are similar to those displayed by age-matched controls (Takesian *et al.*, 2012). A similar outcome has been observed following bilateral hearing loss in developing rats. Membrane excitability is altered at short-term survival intervals, but neurons no longer differ from controls at 1 month postnatal (Rao *et al.*, 2010). Interestingly, serotonin suppresses pyramidal cell discharge, but only at longer survival times. These findings suggest that perceptual deficits that are observed in adulthood are likely due to only a subset of the cellular alterations that have been described following a short survival time.

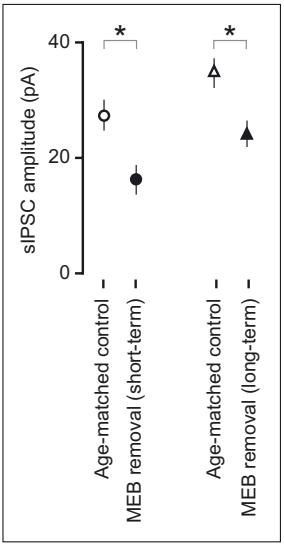

Fig. 6: Following developmental CHL at postnatal day 10, inhibitory synaptic currents remain depressed through adulthood. Spontaneous IPSCs were recorded in auditory cortex pyramidal neurons after 7-12 days of hearing loss (short-term), or 80-100 days of hearing loss (long-term). At both survival times, CHL resulted in smaller IPSCs, as compare to age-matched controls (MEB, middle ear bone). Asterisks indicate a statistically significant change. Adapted from Takesian *et al.* (2012).

SUMMARY

At the qualitative level, it is clear that the consequences of hearing loss on synaptic properties are similar for animals with experimentally induced moderate CHL or SNHL. However, there is not yet sufficient information on synaptic function following mild forms of developmental hearing loss to determine whether its consequences are comparable. Since mild unilateral hearing loss does induce significant changes to CNS coding properties, the likelihood is that mild hearing loss does induce substantive cellular changes. Certainly, our preliminary findings (bilateral earplugs, Fig. 4) are in accord with this conclusion. At the quantitative level, the effects of hearing loss are likely to be of larger magnitude when there is a loss of hair cells and/or spiral ganglion neurons. This is apparent in some (e.g., Fig. 5), but not all, of our measures from auditory cortex. Taken together, these findings suggest that profound changes to central processing may occur even when developmental hearing loss is moderate (CHL) and raises the question whether

transient forms of CHL are equally detrimental to the cellular maturation of central auditory circuits.

REFERENCES

Balakrishnan, V., Becker, M., Lohrke, S., Nothwang, H.G., Guresir, E., and Friauf, E. (**2003**). "Expression and function of chloride transporters during development of inhibitory neurotransmission in the auditory brainstem," J. Neurosci., **23**, 4134-4145.

Brenowitz, S., and Trussell, L.O. (**2001**). "Maturation of synaptic transmission at end-bulb synapses of the cochlear nucleus," J. Neurosci., **21**, 9487-9498.

Chirila, F.V., Rowland, K.C., Thompson, J.M., and Spirou, G.A. (**2007**). "Development of gerbil medial superior olive: integration of temporally delayed excitation and inhibition at physiological temperature," J. Physiol., **584**, 167-190.

Chuhma, N., and Ohmori, H. (**1998**). "Postnatal development of phase-locked high-fidelity synaptic transmission in the medial nucleus of the trapezoid body of the rat," J. Neurosci., **18**, 512-520.

Dorrn, A.L., Yuan, K., Barker, A.J., Schreiner, C.E., and Froemke, R.C. (**2010**). "Developmental sensory experience balances cortical excitation and inhibition," Nature, **465**, 932-936.

Fitzgerald, K.K., and Sanes, D.H. (**2001**). "The development of stimulus coding in the auditory system," in *Physiology of the Ear, 2nd Edition*. Edited by E. Jahn and J. Santos-Sacchi (Singular Publishing, San Diego), pp. 215-240.

Gao, H., and Lu, Y. (**2008**). "Early development of intrinsic and synaptic properties of chicken nucleus laminaris neurons," Neurosci., **153**, 131-143.

Johnson, S.L., Eckrich, T., Kuhn, S., Zampini, V., Franz, C., Ranatunga, K.M., Roberts, T.P., Masetto, S., Knipper, M., Kros, C.J., and Marcotti, W. (**2011**). "Position-dependent patterning of spontaneous action potentials in immature cochlear inner hair cells," Nature Neurosci., **14**, 711-717.

Jones, T.A., Leake, P.A., Snyder, R.L., Stakhovskaya, O., and Bonham, B. (**2007**). "Spontaneous discharge patterns in cochlear spiral ganglion cells before the onset of hearing in cats," J. Neurophys., **98**, 1898-1908.

Kandler, K., and Friauf, E. (**1995**). "Development of glycinergic and glutamatergic synaptic transmission in the auditory brainstem of perinatal rats," J. Neurosci., **15**, 6890-6904.

Kotak, V.C., and Sanes, D.H. (**1997**). "Deafferentation of glutamatergic afferents weakens synaptic strength in the developing auditory system," Eur. J. Neurosci. **9**, 2340-2347.

Kotak, V.C., Fujisawa, S., Leem F.A., Karthikeyan, O., Aoki, C., and Sanes, D.H. (**2005**). "Hearing loss raises excitability in the auditory cortex," J. Neurosci., **25**, 3908-3918.

Kotak, V.C., Breithaupt, A.D., and Sanes, D.H. (**2007a**). "Developmental hearing loss eliminates long-term potentiation in the auditory cortex," Proc. Natl. Acad. Sci. USA, **104**, 3550-3555.

Kotak, V.C., Sadahiro, M., and Fall, C.P. (**2007b**) "Developmental expression of endogenous oscillations and waves in the auditory cortex involves calcium, gap junctions, and GABA," Neurosci., **146**, 1629-1639.

Kotak, V.C., Takesian, A.E., and Sanes, D.H. (**2008**). "Hearing loss prevents the maturation of GABAergic transmission in the auditory cortex," Cerebral Cortex, **18**, 2098-2108.

Kotak, V.C., Péndolam L.M., and Rodríguez-Contreras, A. (**2012**). "Spontaneous activity in the developing gerbil auditory cortex in vivo involves GABAergic transmission," Neurosci., **226**, 130-144.

Kotak, V.C., Takesian, A.E., MacKenzie, P.C., and Sanes, D.H. (**2013**). "Rescue of inhibitory synapse function following developmental hearing loss," PLoS One, **8**, e53438.

Kuba, H., Oichi, Y., and Ohmori, H. (**2010**). "Presynaptic activity regulates Na(+) channel distribution at the axon initial segment," Nature, **465**, 1075-1078.

Magnusson, A.K., Kapfer, C., Grothe, B., and Koch, U. (**2005**). "Maturation of glycinergic inhibition in the gerbil medial superior olive after hearing onset," J. Physiol., **568**, 497-512.

Mowery, T.M., Kotak, V.K., and Sanes, D.H. (**2013**), "Critical period for auditory cortex inhibitory maturation closes by postnatal day 19," Soc. Neurosci. Abs. **43**.

Nakamura, Y., and Takahashi, T. (**2007**). "Developmental changes in potassium currents at the rat calyx of Held presynaptic terminal," J. Physiol., **581**, 1101-1112.

Oswald, A.M., and Reyes, A.D. (**2008**). "Maturation of intrinsic and synaptic properties of layer 2/3 pyramidal neurons in mouse auditory cortex," J. Neurophysiol., **99**, 2998-3008.

Oswald, A.M., and Reyes, A.D. (**2011**). "Development of inhibitory timescales in auditory cortex," Cereb. Cortex, **21**, 1351-1361.

Pinto, J.G., Hornby, K.R., Jones, D.G., and Murphy, K.M. (**2010**). "Developmental changes in GABAergic mechanisms in human visual cortex across the lifespan," Front. Cell. Neurosci., **4**, 16.

Rao, D., Basura, G.J., Roche, J., Daniels, S., Mancilla, J.G., and Manis, P.B. (**2010**). "Hearing loss alters serotonergic modulation of intrinsic excitability in auditory cortex," J. Neurophys., **104**, 2693-2703.

Sanchez, J.T., Wang, Y., Rubel, E.W., and Barria, A. (**2010**). "Development of glutamatergic synaptic transmission in binaural auditory neurons," J. Neurophysiol., **104**, 1774-1789.

Sanes, D.H. (**1993**). "The development of synaptic function and integration in the central auditory system," J. Neurosci., **13**, 2627-2637.

Sanes, D.H., and Walsh, E.J. (**1997**). "Development of Auditory Processing," in *Development of the Auditory System*. Edited by E.W Rubel, A.N. Popper, and R.R. Fay (Springer-Verlag, New York), pp. 271-314.

Sanes, D.H., and Bao, S. (**2009**). "Tuning up the developing auditory CNS," Curr. Opin. Neurobiol., **19**, 188-199.

Sanes, D.H., and Woolley, S.M.N. (**2011**). "A behavioral framework to guide research on central auditory development and plasticity," Neuron, **72**, 912-929.

Sanes, D.H. (**2013**). "Synaptic and cellular consequences of hearing loss," in *Springer Handbook of Auditory Research: Deafness*. Edited by A. Kral, R.R. Fay, and A.N. Popper (Springer-Verlag: New York).

Sarro, E.C., Kotak, V.C., Sanes, D.H., and Aoki, C. (**2008**), "Hearing Loss Alters the Subcellular Distribution of Presynaptic GAD and Postsynaptic GABAA Receptors in the Auditory Cortex," Cereb. Cortex **18**, 2855-2867.

Scott, L.L., Mathews, P.J., and Golding, N.L. (**2005**). "Posthearing developmental refinement of temporal processing in principal neurons of the medial superior olive," J. Neurosci., **25**, 7887-7895.

Takesian, A.E., Kotak, V.C., and Sanes, D.H. (**2010**). "Presynaptic GABA(B) receptors regulate experience-dependent development of inhibitory short-term plasticity," J. Neurosci., **30**, 2716-2727.

Takesian, A.E., Kotak, V.C., and Sanes, D.H. (**2012**). "Age-dependent effect of hearing loss on cortical inhibitory synapse function," J. Neurophys., **107**, 937-947.

Taschenberger, H., and von Gersdorff, H. (**2000**). "Fine-tuning an auditory synapse for speed and fidelity: developmental changes in presynaptic waveform, EPSC kinetics, and synaptic plasticity," J. Neurosci., **20**, 9162-9173.

Tritsch, N.X., Yi, E., Gale, J.E., Glowatzki, E., and Bergles, D.E. (**2007**). "The origin of spontaneous activity in the developing auditory system," Nature, **450**, 50-55.

Tritsch, N.X., Rodríguez-Contreras, A., Crins, T.T., Wang, H.C., Borst, J.G., and Bergles, D.E. (**2010**). "Calcium action potentials in hair cells pattern auditory neuron activity before hearing onset," Nature Neurosci., **13**, 1050-1052.

Vale, C., and Sanes, D.H. (**2000**). "Afferent regulation of inhibitory synaptic transmission in the developing auditory midbrain," J. Neurosci., **20**, 1912-1921.

Vale, C., and Sanes, D.H. (**2002**). "The effect of bilateral deafness on excitatory synaptic strength in the auditory midbrain," Eur. J. Neurosci., **16**, 2394-2404.

Vale, C., Schoorlemmer, J., and Sanes, D.H. (**2003**). "Deafness disrupts chloride transport and inhibitory synaptic transmission," J. Neurosci., **23**, 7516-7524.

Xu, H., Kotak, V.C., and Sanes, D.H. (**2007**). "Conductive hearing loss disrupts synaptic and spike adaptation in developing auditory cortex," J. Neurosci., **27**, 9417-9426.

Xu, H., Kotak, V.C., and Sanes, D.H. (**2010**). "Normal hearing is required for the emergence of long-lasting inhibitory potentiation in cortex," J. Neurosci., **30**, 331-341.