Response of Inferior Colliculus Neurons to Electrical Stimulation of the Auditory Nerve in Neonatally Deafened Cats

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Shepherd, Robert K., Jagir H. Baxi, and Natalie A. Hardie. Response of inferior colliculus neurons to electrical stimulation of the auditory nerve in neonatally deafened cats. J. Neurophysiol. 82: 1363–1380, 1999. Response properties of neurons in the inferior colliculus (IC) were examined in control and profoundly deafened animals to electrical stimulation of the auditory nerve. Seven adult cats were used: two controls; four neonatally deafened (2 bilaterally, 2 unilaterally); and one long-term bilaterally deaf cat. All control cochleae were deafened immediately before recording to avoid electrographic activation of hair cells. Histological analysis of neonatally deafened cochleae showed no evidence of hair cells and a moderate to severe spiral ganglion cell loss, whereas the long-term deaf animal had only 1–2% ganglion cell survival. Under barbiturate anesthesia, severe spiral ganglion cell loss, whereas the long-term deaf animal had only 1–2% ganglion cell survival. Under barbiturate anesthesia, severe spiral ganglion cell loss, whereas the long-term deaf animal had only 1–2% ganglion cell survival.

INTRODUCTION

Removal of auditory input as a result of a sensorineural hearing loss or cochlear ablation leads to atrophic changes within central auditory nuclei. At the level of the cochlear nucleus (CN), deafferentation results in decreased neuronal size (Hashisaki and Rubel 1989; Matsushima et al. 1991; Moore and Kowalchuk 1988; Nordeen et al. 1983; Powell and Erulkar 1962; Trune 1982 and CN volume (Hardie and Shepherd 1999; Lustig et al. 1994; Moore and Kowalchuk 1988; Parks 1979). Furthermore deafferentation before the onset of hearing results in a decrease in CN cell number (Born and Rubel 1985; Hashisaki and Rubel 1989; Nordeen et al. 1983; Tierney et al. 1997; Trune 1982).

Deafferentation at the level of the auditory nerve also leads to morphological changes in more central auditory nuclei. A reduction in neuronal soma area in the trapezoid body has been reported after cochlear ablation (Pasic et al. 1994), and deafness-induced changes in neuron morphology also have been reported in the superior olivary complex and the lateral lemniscus (Powell and Erulkar 1962). In animals deafened neonatally, we recently described a significant reduction in synaptic density within the central nucleus of the inferior colliculus (ICC) of bilaterally deaf compared with normal-hearing animals (Hardie et al. 1998). In contrast, there was no significant difference between normal-hearing and unilaterally deaf animals.

Although loss of auditory input does not appear to result in significant changes in synaptic density in the auditory midbrain of unilaterally deaf animals, neuronal tracing studies have shown a significant increase in the number of CN neurons projecting ipsilaterally to the inferior colliculus (IC) on the unlesioned side of the brain in animals that have undergone unilateral cochlear removal (Moore 1994; Moore and Kowalchuk 1988; Nordeen et al. 1983). In contrast, a bilateral loss does not appear to affect the CN-IC connectivity relative to normal-hearing animals (Moore 1990).

The physiological response properties of neurons within the IC of animals with a unilateral lesion are also significantly altered (Nordeen et al. 1983). For example, in response to acoustic stimulation of the normal cochlea, the relatively small number of loci at which excitatory activity can be recorded in the ipsilateral IC of normal-hearing animals (~30%) increases to ~70% in acute, unilaterally ablated adult animals, whereas cochlear removal in the neonatal period further increases the IC responsiveness to ipsilateral stimulation (80–90%) (McAlpine et al. 1997).

Electrical stimulation of the auditory nerve has been used to study physiological response properties of neurons within the auditory pathway of deaf animals to investigate changes after periods of sensorineural hearing loss. These studies have been focused at the level of the auditory nerve (Parkins and Colombo 1987; Shepherd and Javel 1997), the auditory midbrain (Shirane and Harrison 1991; Snyder et al. 1990, 1991, 1995).
and the auditory cortex (Hartmann et al. 1997). The majority of these studies agree that the basic response properties of electrically evoked single-unit activity appear to be relatively independent of hearing status. For example, neurons typically exhibit a monotonic increase in spike rate and a reduction in both latency and temporal jitter across their dynamic range (Hartmann et al. 1997; Shepherd and Javel 1997; Snyder et al. 1991). In contrast, Shirane and Harrison (1991) have reported little evidence of single-unit activity in the IC after electrical stimulation of the auditory nerve in neonatally deafened animals. These authors concluded that there is minimal afferent input to the auditory midbrain in animals deafened from birth.

We examined the electrophysiological response properties of IC neurons in neonatally deafened cats (studied as adults) to electrical stimulation of the auditory nerve, and compared their response properties with those from acutely deafened control animals. Our recordings have shown that many of the basic response properties are similar across animals with a wide range of auditory experience. However, important differences also were identified, including increased response latencies and temporal jitter and reduced levels of temporal resolution.

**METHODS**

**Animals**

Seven adult cats were used in the present study. Two animals were normal-hearing controls (C; Table 1), the remaining animals were deafened profoundly by administration of ototoxic drugs. Four were deafened neonatally—two bilaterally (BD) and two unilaterally (UD). One animal (LT-17), deafened as a juvenile, had a profound bilateral hearing loss of 93-mo duration. The normal-hearing animals and the normal cochleae of the unilaterally deaf animals were deafened immediately before single-unit recording to eliminate contamination by electrophonic activation of inner hair cells (Moxon 1971; Shepherd and Javel 1997). For simplicity, these cochleae are referred to as controls.

**Deafening procedures**

With the exception of the two control animals and LT-17, animals were deafened at 10 days after birth (DAB). Bilateral deafness was induced via a single coadministration of kanamycin (KA; kanamycin monosulphate) and ethacrynic acid (EA; ethacrynate sodium). Kittens were deafened at 10 days after birth (DAB). Bilateral deafness was achieved by administration of ototoxic drugs. Four were deafened neonatally—two bilaterally (BD) and two unilaterally (UD). One animal (LT-17), deafened as a juvenile, had a profound bilateral hearing loss of 93-mo duration. The normal-hearing animals and the normal cochleae of the unilaterally deaf animals were deafened immediately before single-unit recording to eliminate contamination by electrophonic activation of inner hair cells (Moxon 1971; Shepherd and Javel 1997). For simplicity, these cochleae are referred to as controls.

**TABLE 1. Summary of hearing status and duration of deafness for each animal in this study**

<table>
<thead>
<tr>
<th>Animal*</th>
<th>Hearing Status†</th>
<th>Duration of Deafness, mo</th>
</tr>
</thead>
<tbody>
<tr>
<td>C-1</td>
<td>Control (7; 12)</td>
<td>—</td>
</tr>
<tr>
<td>C-2</td>
<td>Control (22; 22)</td>
<td>—</td>
</tr>
<tr>
<td>BD-5</td>
<td>Bilaterally deaf (&gt;92; &gt;92)</td>
<td>12</td>
</tr>
<tr>
<td>BD-6</td>
<td>Bilaterally deaf (&gt;92; &gt;92)</td>
<td>13</td>
</tr>
<tr>
<td>UD-10</td>
<td>Unilaterally deaf (&gt;92; 7)</td>
<td>12</td>
</tr>
<tr>
<td>UD-11</td>
<td>Unilaterally deaf (&gt;92; 12)</td>
<td>12</td>
</tr>
<tr>
<td>LT-17</td>
<td>Bilaterally deaf (&gt;92; &gt;92)</td>
<td>93</td>
</tr>
</tbody>
</table>

* The nomenclature used to identify these animals is based on that used by Hardie and Shepherd (1999). C, control; BD, bilaterally deaf; UD, unilaterally deaf; LT, long-term deaf. † Hearing status determined by click-evoked auditory brain stem response thresholds. Thresholds, in DB p.e. SPL, are given in parentheses for both ears. For unilaterally deaf cats, thresholds for the deafened ear precedes that for the normally functioning ear.

**Stimulating electrodes**

A bipolar electrode array was implanted ipsilaterally to the IC recording site (Fig. 1). In unilaterally deaf animals, this electrode was implanted in the control cochlea. The array consisted of two 0.3-mm-wide platinum (Pt) ring-shaped electrodes of 0.4–0.5 mm diam on a Silastic carrier. The interelectrode separation was 0.45 mm. A second electrode array, implanted in the contralateral cochlea, consisted of eight Pt electrodes. In unilaterally deaf animals, this array was inserted into the deafened cochlea. Its geometry was identical to the array described above. To study spatial selectivity, both the most apical electrode pair (1–2) and a more basal pair (6–7 or 5–7) were used to evoke neural activity.

**Surgical preparation**

Anesthesia was induced with an intraperitoneal injection of ketamine hydrochloride (20 mg/kg) and xylazine (3.8 mg/kg) and maintained with sodium pentobarbital (35 mg/kg iv), administered periodically to maintain the animal a-reflexic. Atropine sulfate (0.05 mg/kg im) and dexamethasone (0.025 mg/kg im) were administered daily to minimize mucosal secretions and brain edema respectively. A tracheal cannulation was performed and the animal’s head was secured in normal saline, was injected subcutaneously (300 mg/kg). Approximately 30 min later, EA diluted in normal saline to 1 mg/ml was infused intravenously during a 30-s period at a dose of 25 mg/kg (Shepherd and Martin 1995). Animal LT-17 was deafened as a juvenile using the same procedure.

A unilateral hearing loss was induced by neomycin perfusion of the cochlea (Hardie and Shepherd 1999). The animals were anesthetized with an intraperitoneal injection of ketamine hydrochloride (20 mg/kg) and xylazine (3.8 mg/kg). Using sterile surgical techniques the auditory bulla was exposed and the round and oval windows incised. Approximately 2.5 ml of neomycin sulfate (10 mg/ml in normal saline) was infused through the cochlea with gentle aspiration at the oval window. Both windows were closed with muscle and the wounds sutured. All control cochleae were deafened just before the acute physiological experiment using the same technique.

**FIG. 1. Schema of the experimental setup. Stimulating electrodes were implanted into both cochleae. Localized sectors of auditory nerve were stimulated electrically using biphasic current pulses. Cochlea ipsilateral to the inferior colliculus (IC) was stimulated using the apical electrode pair (1–2) only. Contralateral cochlea was stimulated with the apical pair (1–2) and a more basal pair (5–7 or 6–8). In unilaterally deaf animals, the deafened cochlea was contralateral to the IC from which recordings were made. Unit activity was recorded using an electrode advanced at 45° to the sagittal plane.**
stereotaxic apparatus (Trent-Wells). The animal’s core body temperature was maintained at 38 ± 1°C by a feedback electric blanket. Respiration rate and endtidal CO$_2$ were monitored throughout the experiment and kept within normal limits (respiration rate: 10–18/min; endtidal CO$_2$: 2–5%).

Both bullae were exposed surgically, and the round window was incised gently. The electrode array was inserted 6 mm into the scala tympani, and the round window covered with muscle. The occipital cerebral cortex was aspirated to expose the dorsal surface of the IC. The tentorium was drilled to expose the complete dorsolateral surface of the IC and the exposure was covered with warm paraffin oil.

**Electrical stimuli**

Electrical stimuli were generated by an optically isolated current-source stimulator under computer control. The standard stimulus consisted of 100 μs/phase charge-balanced biphasic current pulses presented at 5 pulses/s (pps) for a total of 50 presentations at a given current level. As this stimulus consisted of a single current pulse per repetition period, no attempt was made to differentiate sustained versus onset units. To study temporal response properties, stimulus rate was varied systematically from 5 to 200 pps. For all experiments, the apical electrode of each bipolar pair was cathodic during the first phase of the stimulus.

**Evoked potentials**

The hearing status of all animals was evaluated using auditory brain stem responses (ABRs). Animals were sedated with a mixture of ketamine hydrochloride (20 mg/kg) and xylazine (3.8 mg/kg). Evoked potentials were recorded in an electrically isolated, sound-attenuated Faraday room. Rarefaction clicks (100 μs) were produced via a Richard Allen DT-20 loudspeaker placed 0.1 m from the pinna. An ear mold compound (Otoform) was used to plug the contralateral ear. In the unilaterally deaf animals, white noise (HP 8057A) was presented to the normal ear via a closed acoustic-delivery system during recording (Hardie and Shepherd 1999). ABRs were recorded differentially using subcutaneous electrodes (vertex + ve; neck - ve; thorax ground). Responses were amplified (10$^5$; DAM 5A, WPI) and bandpass filtered (150 Hz to 3 kHz; Krohn Hite 3750). The output was passed through a 10 bit A/D converter and sampled at 20 kHz for 12.5 ms after stimulus onset. Thirty clicks were presented every second and 250 responses were averaged. Threshold for normal ears was <32 dB peak-equivalent (pe) sound pressure level (SPL), while all deafened ears had thresholds >92 dB pe SPL. The hearing status of all deafened animals was determined by 1 wk before the experiment. The hearing status of all deafened animals remained unchanged during this period.

Electrically evoked ABRs (EABRs) were recorded before single-unit recording to confirm that current pulses delivered to the electrode array were stimulating the auditory nerve. Due to the tapered nature of the basal turn of the cochlea, basal electrodes consistently evoke higher thresholds than more apical electrodes. To ensure a useful dynamic range, the basal-most bipolar pair of the array having an EABR threshold of <1.0 mA was selected. Although this was typically electrode pair 6–8, in some animals pair 5–7 was selected. A detailed analysis of the EABRs recorded in these animals is given elsewhere (Hardie and Shepherd 1999).

**Single-unit recordings**

An overview of the recording procedure is illustrated in Fig. 1. Single-unit recordings were made extracellularly using tungsten microelectrodes (impedances: 0.8–1.2 MΩ at 1 kHz). The microelectrodes were located on the IC surface under vision and advanced using a remote-controlled stepping motor (Narashigi). Agar (3%) was used to provide mechanical stability. Penetrations were offset from the sagittal plane by 45° so trajectories were approximately orthogonal to isofrequency laminae (Brown et al. 1992; Semple and Aitkin 1979; Snyder et al. 1990). This ensured that the recording electrode passed through the lateral and the central nuclei of the IC (Morest and Olver 1984).

Single-unit activity was differentially amplified (DAM-5A, WPI), the stimulus artifact eliminated using a sample and hold circuit, and the signal band-pass filtered (150 Hz-3kHz; Krohne-Hite 3750). Output from the filter was displayed on an oscilloscope (Tektronix 465), and its triggering was adjusted to discriminate the leading edge of the action potential. The resultant trigger pulse generated by the oscilloscope was fed to the programmable clock of a PDP11–34 computer to register times of occurrence of discharges with a resolution of 10 μs. The PDP11–34 was also responsible for control and generation of all electrical stimuli.

Unit activity was isolated via an electrical search stimulus delivered randomly to either the ipsi- or contralateral electrode array. Once isolated, spontaneous rate was determined. Rate-intensity functions were recorded in response to 5 pps current pulses presented nonsimultaneously to the ipsilateral and contralateral electrodes. Poststimulus time (PSTH), period (PH), and interspike interval histograms, and rate-intensity functions were displayed dynamically during recording. Mean first driven spike latency and its standard deviation were evaluated from the PSTH. Standard deviation of latency was used as a measure of temporal jitter. Rate-intensity functions were normalized to the number of stimulus presentations to create response probability-intensity (input-output) functions, such that a response probability ($P$) of 1.0 represented one spike in response to each pulse presentation. Threshold and saturation were calculated from the input-output functions using a least-squares fit of a saturating Gaussian function (Sachs and Abbas 1974).

Threshold and saturation were defined as the current level for which $P = 0.1$ and 0.9, respectively. Dynamic range was defined as the change in stimulus intensity from threshold to saturation. Response latencies were fitted with a least-squares double-exponential function:

$$
\text{Latency} = k_1 \exp(-k_2 P) + k_3 \exp(-k_4 P); \quad \text{where } I \text{ is the current intensity, and } k_1, k_2, k_3, k_4 \text{ are constants.}
$$

This fit most accurately modeled the initial rapid decrease in latency at near-threshold current levels, followed by a smaller asymptotic decrease near saturation. This equation was used to obtain response latencies at threshold and saturation current levels. MATLAB (MathWorks) software was used for all curve-fitting procedures. An $r^2$ value (residual sum of squares) of each curve fit was used as an indicator of variation; all fits where $r^2 < 0.95$ were rejected (<10% of the data).

Spatial tuning curves (STCs) were constructed by plotting single-unit thresholds as a function of recording depth (Snyder et al. 1990). Response latencies, thresholds ($P = 0.1$) and dynamic ranges for neonatally deafened animals were compared with corresponding response measures from control animals using the t-test or the Mann-Whitney Rank Sum test where data were not normally distributed. Comparisons of ipsilateral and contralateral response properties were made on paired data only.

**Analysis of temporal response properties**

A group of 76 ICC neurons from both control and neonatally deafened animals were stimulated at rates of 5–200 pps to study temporal response properties. Using 100 μs/phase biphasic current pulses, rate-intensity functions were recorded over each neuron’s dynamic range in response to increasing stimulus rates until the neuron clearly failed to reach saturation ($P = 0.9$). Each rate-intensity function was converted to an input-output function and fitted using a saturating Gaussian function.
**RESULTS**

The hearing status of all animals used in this study is summarized in Table 1. A detailed analysis of morphological changes within the cochlea and cochlear nucleus as a function of hearing status is given elsewhere (Hardie and Shepherd 1999). Briefly, the cochlea of both control animals showed complete inner and outer hair cell loss throughout all turns. In contrast, there was no evidence of loss of spiral ganglion cells. The control cochlea of each unilaterally deaf animal exhibited similar histological features. All neonatally deafened animals were studied at ~12 mo after deafening. Histological analyses of their cochleae showed no evidence of consistent inner or outer hair cell survival (isolated inner hair cells occasionally were observed in the apical turn of some animals). These cochleae also displayed large reductions in spiral ganglion cell density (~15% of normal) (Hardie and Shepherd 1999). The cochlea from animal LT-17, deafened for a period of 93 mo, showed no evidence of surviving hair cells and extensive reductions in the ganglion cell density throughout the cochleae (1–2% of normal).

**Single-unit results**

This analysis is based on recordings from 419 discriminated single-unit responses (control: 128; bilaterally deaf: 118; unilaterally deaf: 106; long-term deaf: 67). Representative responses from a control and a unilaterally deaf animal are illustrated in Fig. 2. Both responses consist of several traces evoked by a fixed current amplitude stimulus. The associated PSTHs for these neurons also are shown. Note that the neuron recorded from C-I was located superficially (recording depth, 1.520 µm), and therefore probably was located within the LN. Long-latency responses were a feature of units isolated superficially (see Spatial distribution of unit responses).

**Spontaneous activity as a function of hearing status**

The distribution of spontaneous activity as a function of hearing status is illustrated in Fig. 3. It is important to recall that the two control animals, as well as the control cochlea of unilaterally deaf animals, were deafened just before the experiment. Therefore the level of spontaneous activity reported here does not reflect levels that would be expected in normal-hearing animals.

Mean spontaneous activity across all animals was <8 spikes/s. Fifty-nine percent of cells isolated from the control animals showed no spontaneous activity. Of the remaining cells, the majority (38%) displayed spontaneous activity ≤5 spikes/s. In unilaterally deaf animals, similar proportions were observed, with 60% of units displaying no spontaneous activity and 28% of units displaying spontaneous activity ≤5 spikes/s. Bilaterally deaf animals exhibited a smaller proportion of cells with no spontaneous activity (bilaterally deaf, 25%; long-term deaf, 43%), and a larger proportion of cells with spontaneous rates ≤5 spikes/s (bilaterally deaf, 45%; long-term deaf, 39%). In addition, 31% of neurons recorded from the bilaterally deaf animals and 18% of neurons in the long-term deaf animal exhibited spontaneous rates >5 spikes/s. A statistical comparison showed that bilaterally deaf animals exhibited a significantly higher median spontaneous rate (1.35, interquartile range 0.1–7.8 spikes/s) compared with the acutely deafened control animals (0.0, 0.0–0.2 spikes/s; P < 0.0001; Mann-Whitney rank sum test). The long-term deaf animal also exhibited a significant increase in spontaneous activity (0.10, 0.0–1.7 spikes/s) when compared with the control animals (P = 0.008). In contrast, there was no statistically significant difference between the unilaterally deaf (0.00, 0.0–0.8 spikes/s) and control animals (P = 0.4281).

**Binaural response properties**

The distribution of neuron type based on their binaural response properties is illustrated in Fig. 4. Six binaural response types were observed. Control and neonatally deafened animals showed very similar distributions, with ~80% of cells from all groups showing an excitatory response to contralateral stimulation and ~75% showing an excitatory response to ipsilateral stimulation. Units excited by stimulation of both cochleae, EE units, represented 60–70% of all neurons encountered in these groups. Although the proportion of EE units was lower in LT-17 (50% of the total neural population), the high proportion of EO units in this animal (45%) possibly included a number of cells that could not be driven with ipsilateral stimulation at maximum current levels due to elevated thresholds (Table 2). A small proportion of neurons were excited by ipsilateral stimulation but unresponsive to contralateral stimulation (10% in control/unilaterally deaf; 5% in bilaterally deaf). The small proportion of cells exhibiting inhibitory activity (EI, IE, and II cells) in these animals reflects the relatively low levels of spontaneous activity in the IC.

**General properties of excitatory responses**

Figure 5 illustrates the fine temporal response properties of three short latency (<10 ms) excitatory responses recorded from a control (C-I), neonatally deaf (BD-5), and long-term deaf (LT-17) animal. Responses were elicited by stimulation of the cochlea contralateral to the IC. Responses to ipsilateral stimulation showed no qualitative difference.

Response properties across animals with varying degrees of cochlear pathology appeared quite similar. Neurons were well synchronized to the current pulse and typically showed a monotonic increase in spike probability with increasing stimulus current. Latency and temporal jitter generally decreased monotonically with increasing current. While at saturation, each pulse generally elicited one spike, at higher intensities
many neurons exhibited two and sometimes three spikes in response to each current pulse (Fig. 5).

Although single-unit activity could be elicited readily from the IC of neonatally deafened animals, there was a reduction in the number of cells isolated per millimeter of recording track compared with control animals. This reduction was generally not statistically significant (Table 3). This table also highlights the dominance of contralateral compared with ipsilateral input to the IC.

Typical examples of single-unit input-output functions, fitted with saturating Gaussian functions, are illustrated in Fig. 6. Their general characteristics were not affected by hearing status. Although in this example the response from the control animal exhibited the lowest threshold, this was highly dependent on the recording site within the IC (see Spatial distribution of unit responses). A slight increase in dynamic range in neonatally deafened compared with control cochleae was often observed. This is depicted in Fig. 6 as a reduction in the gradient of the input-output function.

Neurons also typically showed a systematic reduction in latency with increasing stimulus intensity irrespective of hearing status (Fig. 6). Note that neurons BD-6–57 and LT-17–9, both of which exhibited short-latency responses (<10 ms at P ≥ 0.9), were recorded at depths of ~4,000 μm from the IC surface and therefore were considered to be within the central nucleus. Longer latency neurons (10–25 ms at P ≥ 0.9) were isolated from the superficial region of each track, illustrated here by neuron C-2–4 (recorded at a depth of ~750 μm).

Approximately 15% of IC neurons isolated from control and neonatally deaf animals displayed nonmonotonic behavior, however this proportion was reduced to 4% in the long-term deaf animal. Similar proportions of nonmonotonic neurons were seen for both ipsi- and contralateral stimulation, although a cell exhibiting nonmonotonic behavior to stimulation from one cochlea did not necessarily show a similar pattern in response to stimulation from the other cochlea. Input-output functions for nonmonotonic units almost always reached saturation before the spike probability reduced (typically to P = 0.4–0.6, but sometimes less; Fig. 7).
Inhibitory activity

All cells exhibiting spontaneous activity showed inhibition in response to electrical stimulation of the auditory nerve (Fig. 8). Threshold for suppression of spontaneous activity was generally lower than the threshold for excitatory activity. Moreover, the period of inhibition increased with increasing stimulus intensity, ranging from ≈30 ms near threshold, to periods >100 ms.

Spatial distribution of unit responses

Two basic response properties were identified when recording from the superficial layers of the IC (recording depths ≤1,600 μm). Figure 9 illustrates a typical example of each response recorded from two neurons isolated from a control animal. Both examples were evoked in response to contralateral stimulation using the standard stimulus paradigm (100 μs/phase biphasic pulses at 5 pps). The first response type (C-1–50, Fig. 9) consisted of a long-latency (10–25 ms) excitatory response with relatively large temporal jitter and narrow first-spike dynamic range. Spike latency decreased with increasing stimulus intensity. These cells often showed multispiking (usually 3–4 spikes but up to 7 spikes per pulse), and many displayed strong nonmonotonic behavior. Furthermore these units were more strongly driven by stimulation of the contralateral cochlea compared with the ipsilateral cochlea. The second response type (C-1–12, Fig. 9), which was not frequently encountered, consisted of a highly unsynchronized excitatory response. Although spike probability increased with current level, there was no clear response peak. These responses could be obtained by both ipsi- and contralateral stimulation. Both response properties were observed across all groups except the long-term deaf animal LT-17 (see following text). Given that these responses were only recorded superficially, it is reasonable to conclude that they reflect activity recorded from neurons within the LN.

At deeper recording sites (>1,600 μm), cells responded with the short-latency excitatory response (5–8 ms at saturation) described previously (e.g., Figs. 5 and 6). This short-latency activity appears to be characteristic of neurons within the ICC. The great majority of responses recorded in the present study were of this type.

The extracellular traces depicted in Fig. 2 emphasize the latency difference seen between neurons within the LN and the ICC. The latency of excitatory LN neurons exhibited an orderly decrease from ≈25 to <15 ms with increasing recording depth. At deeper recording sites, response latencies were typically 5–8 ms at saturation and showed no further reduction with recording depth. Figure 10 illustrates the response latency at saturation plotted as a function of recording depth for four animals in this study. This spatial distribution of latencies was evident for all animals in the present study with the exception of the long-term deaf animal LT-17. In this animal, fewer cells were isolated during the course of any one trajectory through the IC, and driven activity was observed only at depths >2,500 μm. All cells isolated in LT-17 exhibited short-latency excita-
tory responses and hence were considered to be exclusively ICC neurons.

**ICC response latencies as a function of hearing status**

As shown in Fig. 6, the response properties of units isolated within the ICC showed a systematic reduction in spike latency as stimulus current was increased. This reduction was evident for both ipsilateral and contralateral stimulation. Figure 11 illustrates the median latency at threshold, mddynamic range, and saturation. In general, neonatally deafened animals had longer latencies than controls; latencies from the long-term deafened animal were generally significantly longer. Similar increases were observed for stimulation of the neonatally deafened cochlea in unilaterally deafened animals. In contrast, the latency of ICC neurons to stimulation of the control cochlea in these animals showed no significant difference when compared with control animals (Fig. 11).

Ipsilateral-contralateral latency differences for ICC units were analyzed at saturation ($P < 0.9$) using paired data only. Figure 12 illustrates the median ipsilateral-contralateral latency difference for each group expressed as a percentage of the contralateral latency. Neural response latency was, in general, significantly shorter in response to ipsilateral compared with contralateral stimulation. Control animals exhibited an ipsilateral-contralateral latency difference of $27.5\%$ ($n = 50$). That is, cells within the ICC show a $7.5\%$ reduction in median latency when the cochlea ipsilateral to the IC was stimulated compared with stimulation of the contralateral cochlea. This difference was highly statistically significant ($P < 0.0001$; Wilcoxon signed rank test). The neonatally deafened animals also showed significant ipsilateral-contralateral differences (unilaterally deaf, $-8.7\%, n = 52$; bilaterally deaf, $-5.4\%, n = 50$; $P = 0.004$). In contrast, the long-term deaf animal showed only a marginally shorter latency for ipsilateral compared with contralateral stimulation. This difference was not significant ($-0.6\%, n = 21$; $P = 0.980$).

Many neurons could be excited by stimulation of two distinct sectors of auditory nerve corresponding to the apical and basal bipolar electrodes in the contralateral cochlea. Figure 13 compares the response latency of 125 neurons from both the LN and ICC evoked by stimulation of the apical electrode pair (1–2; ~6 mm from the round window) to that evoked by the basal electrode pair (5–7 or 6–8) located just inside the round window.

### TABLE 2. Mean thresholds for ipsilateral and contralateral stimulation

<table>
<thead>
<tr>
<th>Hearing Status</th>
<th>Animal</th>
<th>Ipsilateral Threshold</th>
<th>Contralateral Threshold</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>C-1</td>
<td>1.57 ± 0.29</td>
<td>1.24 ± 0.06</td>
</tr>
<tr>
<td></td>
<td>C-2</td>
<td>1.03 ± 0.10</td>
<td>1.05 ± 0.12</td>
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<tr>
<td>Bilaterally deaf</td>
<td>BD-5</td>
<td>0.98 ± 0.11</td>
<td>0.79 ± 0.11</td>
</tr>
<tr>
<td></td>
<td>BD-6</td>
<td>1.22 ± 0.08</td>
<td>1.07 ± 0.05</td>
</tr>
<tr>
<td>Unilaterally deaf*</td>
<td>UD-10</td>
<td>1.03 ± 0.09</td>
<td>0.83 ± 0.08</td>
</tr>
<tr>
<td></td>
<td>UD-11</td>
<td>1.18 ± 0.10</td>
<td>0.95 ± 0.10</td>
</tr>
<tr>
<td>Long-term deaf</td>
<td>LT-17</td>
<td>1.86 ± 0.08</td>
<td>1.47 ± 0.10</td>
</tr>
</tbody>
</table>

Values are means ± SE in milliamps. *Unilaterally deaf animals were deafened on the side contralateral to the inferior colliculus (IC) recording site.
These data, obtained at saturation, show that there is no latency difference between stimulation of different sectors of auditory nerve. The regression line, representing all the data, had a slope of 1.01 and passes almost through the origin. Statistically, a neuron’s response latency to stimulation of the apical electrode pair was highly correlated with its latency to stimulation of the basal pair ($P < 0.0001$; linear regression analysis).

**Temporal jitter**

Temporal jitter decreased across each neuron’s dynamic range regardless of the animal’s hearing status. A quantitative comparison of temporal jitter as a function of hearing status was made from a total of 204 cells (Table 4). Results were based on jitter measured at the lowest current level required to achieve saturation. Only ICC neurons were included in the analysis. There was an increase in temporal jitter among neonatally deafened animals compared with the controls, although this increase was not always statistically significant (Table 4). Moreover, temporal jitter was consistently greater for contralateral versus ipsilateral stimulation.

Temporal jitter also increased with spike latency. Figure 14 illustrates temporal jitter plotted as a function of spike latency for a total of 170 neurons from both control and neonatally deafened animals. These data, obtained at saturation, represent activity evoked via stimulation of the contralateral cochlea only, and include long-latency responses from the LN. A linear regression line was fitted to the data, and is illustrated in the figure along with its $r^2$ value. Temporal jitter was highly correlated with latency ($P < 0.0001$; linear regression analysis).

**Response thresholds**

Single-unit thresholds generally ranged between 0.3 and 3.0 mA with mean thresholds for ipsilateral stimulation generally higher than for contralateral stimulation. This was even the case for unilaterally deaf animals where thresholds for the control cochlea (ipsilateral) were higher than thresholds for the deafened (contralateral) cochlea (Table 2).

Figure 15 shows examples of STCs, illustrating the variation in single-unit threshold as a function of recording depth for a single track through the IC. In all cases, neural activity was evoked by stimulation of the contralateral cochlea. Responses (○) evoked by the apical electrode pair (1–2) were located ~6 mm from the round window (~12- to 15-kHz region of the cochlea) (Liberman 1982), whereas response thresholds (●) to the basal electrodes were located just inside the round window (~30-kHz region). The examples illustrated here are based on electrode tracks in which a sufficient number of single units were recorded in order that STCs could be constructed. This was not the case in every track, and multiunit recordings were not made in this

TABLE 3. Mean number of single-unit responses isolated per millimeter of recording track as a function of hearing status

<table>
<thead>
<tr>
<th>Hearing status</th>
<th>Units/mm</th>
<th>n</th>
<th>$P^*$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Contralateral stimulation</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>2.78 ± 0.34</td>
<td>10</td>
<td>0.212 NS</td>
</tr>
<tr>
<td>Bilaterally deaf</td>
<td>2.29 ± 0.20</td>
<td>12</td>
<td>0.142 NS</td>
</tr>
<tr>
<td>Unilaterally deaf†</td>
<td>2.18 ± 0.19</td>
<td>10</td>
<td>0.914 NS</td>
</tr>
<tr>
<td>Long-term deaf</td>
<td>1.92 ± 0.49</td>
<td>6</td>
<td>0.033</td>
</tr>
<tr>
<td>Ipsilateral stimulation</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>1.63 ± 0.30</td>
<td>10</td>
<td>0.388 NS</td>
</tr>
<tr>
<td>Bilaterally deaf</td>
<td>1.29 ± 0.21</td>
<td>12</td>
<td>0.120 NS</td>
</tr>
<tr>
<td>Unilaterally deaf†</td>
<td>1.59 ± 0.15</td>
<td>10</td>
<td>0.061</td>
</tr>
<tr>
<td>Long-term deaf</td>
<td>0.59 ± 0.18</td>
<td>6</td>
<td>0.946</td>
</tr>
</tbody>
</table>

Recording track includes both the superficial lateral and central IC (LN and ICC). Units are means ± SE. * $t$-test. Data compared with controls. † Unilaterally deaf animals were deafened on the side contralateral to the IC recording site.
However, recordings of thresholds of single units are unambiguous, and the systematic change in threshold with recording depth as illustrated by the examples in Fig. 15 was evident in most tracks from all animals: that is, the lowest threshold for stimulation of the basal electrode pair occurred at a deeper penetration depth than did the lowest threshold for stimulation of the apical pair. However, although there was evidence of spatial tuning, the distance (in μm) between threshold minima (indicated by 1 in Fig. 15) for electrodes 1–2 and the basal electrode pair was reduced in neonatally deafened compared with control animals. We therefore refer to the spatial tuning evident in the neonatally deafened animals as “rudimentary.” Also evident from Fig. 15, minimum threshold for the more basal electrode pair is consistently higher than that associated with electrode pair 1–2 due to the tapered nature of the scala tympani. Bipolar electrodes in the lower basal turn are further from their target neural elements, resulting in elevated thresholds compared with electrodes located more apicalward (Shepherd et al. 1993).

Dynamic range

There was no significant difference in dynamic range of LN neurons when compared with neurons isolated within ICC ($P = 0.283$, t-test, $n = 212$). ICC single-unit dynamic ranges varied between 0.02 and 7 dB, with dynamic ranges typically falling between 1.5 and 3.0 dB. In general, neonatally deafened animals exhibited wider dynamic ranges than controls. In a number of cases, this increase was statistically significant (Table 5). In both controls and the long-term deaf animal, dynamic ranges were wider for contralateral compared with ipsilateral stimulation (Table 5), although this was not statistically significant. For unilaterally deaf animals, this trend was reversed with dynamic ranges to ipsilateral stimulation wider than for contralateral stimulation. In neonatally, bilaterally deaf animals, there was little difference in the dynamic range of ipsi- versus contralateral responses.

Temporal response properties

Figure 16 illustrates two input-output functions to increasing rates of stimulation. Although the neuron recorded from the control animal was able to reach a response probability of 1 (i.e., a spike in response to each current pulse) at stimulus rates of ≤120 pps, the neuron isolated from the neonatal bilaterally deaf animal reached saturation at rates ≤40 pps. When stimulated at a rate of ≥50 pps, this neuron exhibited maximum response probabilities <1. Although there was wide variability among neurons, cells from control and unilaterally deaf animals consistently showed an ability to respond at high levels of entrainment at stimulus rates significantly higher than those from the neonatally bilaterally deaf animals (Fig. 17).

Figure 18 illustrates a negative correlation between a unit’s first spike latency and its maximum temporal resolution to electrical stimulation. Although the coefficient of determination was low ($r^2 = 0.263$), the correlation was statistically significant ($P < 0.0001$).
DISCUSSION

Many of the single-unit response properties in the present study showed similar characteristics despite a wide range of cochlear pathology. The great majority of cells exhibited a monotonic increase in spike probability with a concomitant reduction in first spike latency and temporal jitter as a function of stimulus intensity. In addition, the majority of neurons across the four groups of animals showed evidence of multiple spiking to a single current pulse as stimulus intensities approached saturation. Finally, evidence of a cochleotopic organization in the ICC of all animals suggests that such a structure can be laid down, at least in a rudimentary form, in the absence of auditory experience.

However, a number of important differences also were identified. First spike latency increased with hearing loss, and although limited to one animal, greater increases were associated with an extended duration of deafness. Deafness-related increases in temporal jitter and dynamic range were also evident. Finally, temporal resolution was significantly reduced in neonatal bilaterally deaf animals compared with both neonatal unilaterally deaf and acutely deafened control animals.

Changes in the cochlear periphery after a sensorineural hearing loss

A number of important morphological changes occur after loss of the sensory epithelium. First, there is a rapid and extensive loss of the unmyelinated sector of the peripheral dendrites within the organ of Corti (Terayama et al. 1977). A more gradual degeneration of the auditory nerve fibers (ANFs) follows, resulting in a significant reduction in the packing density of both the myelinated portion of the peripheral dendrites and their cell bodies (Hardie and Shepherd 1999; Nadol et al. 1989; Shepherd and Javel 1997). The degenerative process also results in de-myelination of residual spiral ganglion soma and their peripheral processes (Spoendlin 1984). Antero-grade degeneration of ANFs is an ongoing process, eventually resulting in very small numbers of surviving ganglion cells in

FIG. 8. Representative period histograms illustrating inhibition of spontaneous activity in the IC in response to electrical stimulation of the auditory nerve for control, bilaterally deaf, and unilaterally deaf animals. Common stimulus (100 μs/phase biphasic current pulses at 5 pps) delivered to electrode pair 1–2 of the contralateral cochlea was used in each example illustrated here. Each histogram consists of 50 repetitions. Stimulus current increases down each column.

FIG. 9. Period histograms illustrating the 2 typical response types in the superficial layers (<1.600 μm) of the IC. First response type (C–1–50) consisted of a relatively long-latency (~25 ms) excitatory response with extensive multispiking and a relatively narrow dynamic range. Second type (C–1–12) consisted of a very poorly synchronized excitatory response, with no clear period histogram (PH) peak and no clear relationship between spike number and stimulus level. Note the difference in time base for the 2 sets of PHs. See Fig. 8 for stimulus parameter details.
Spontaneous activity

Loss of the sensory epithelium and the subsequent degeneration of spiral ganglion cells leads to changes in the physiological response properties of residual ANFs including a reduction in the level of spontaneous activity (Liberman and Kiang 1978), although this activity may increase slightly in animals deafened for long periods of time (Shepherd and Javel 1997). Our present results show that prolonged periods of bilateral deafness leads to a slight but significant increase in the level of spontaneous activity at the level of the IC compared with control animals. In contrast, unilaterally deaf animals showed no significant change in the levels of spontaneous activity compared with the controls. However, these data must be interpreted cautiously as the control animals in the present study were acutely deafened. Their levels of spontaneous activity were much lower than that reported by Bock and Webster (1974) in cats with normal acoustic sensitivity under similar conditions of anesthesia. Finally, spontaneous activity showed no evidence of the bursting activity seen in ANFs of sensorineural deafened cats (Shepherd and Javel 1997).

Binaural response properties

In normal-hearing animals, almost all neurons within the ICC are excited by acoustic stimulation of the contralateral ear, whereas ipsilateral stimuli have excitatory influences on <40% of units (Aitkin 1985; Semple and Aitkin 1979; Semple and Kitzes 1985). However, a unilateral hearing loss can result in an increased ipsilateral responsiveness. In normal hearing ferrets, for example, 33% of recording loci in the IC are excited by acoustic stimulation of the ipsilateral ear (McAlpine et al. 1997). This rises to 70% in animals acutely deafened unilaterally by cochlea ablation as adults and to 80–90% in animals deafened as neonates.

In the present study, the most common cell type, regardless of hearing status, was the EE neuron. With the exception of the long-term deaf animal, stimulation of the ipsilateral cochlea resulted in an excitatory response in >75% of all recorded neurons. Thus a unilateral, sensorineural hearing loss initiated early in development had apparently little effect on the responsiveness of the ipsilateral IC compared with control animals in this study because even in the controls almost as many cells responded to ipsilateral as to contralateral stimulation. Furthermore there was no significant increase in the number of responsive units/millimeter of recording track in the IC ipsilateral to the control ear in the unilaterally deaf animals compared with control animals.

It would appear that an electrical stimulus is very effective in evoking an excitatory response in the ipsilateral IC, regardless of hearing status. In trying to account for the general increase in ipsilateral excitability in these animals compared with previous studies using acoustic stimulation, it is difficult to separate the effects of the nature of the stimulus from the effects of deafening. In a study of the effects of electrical
stimulation on binaural response properties in the IC, Lithgow and Clark (1995) compared the ratio of unit types in the IC in response to electrical stimulation of one cochlea, which was acutely deafened, and acoustic stimulation of the other cochlea. They found an increase in the proportion of units driven when the ipsilateral cochlea was stimulated acoustically and the contralateral cochlea was deafened and electrically stimulated. The authors suggested that this change in the ratio of excitatory and inhibitory inputs to the IC was partly the result of a change in the balance of spontaneous activity in the IC resulting from deafening the contralateral cochlea. Spontaneous activity derived from the contralateral side may act to inhibit afferent activity in the IC from the ipsilateral side. Disruption of this spontaneous activity after deafening the contralateral cochlea could result in an increase in the efficiency of the normally weak ipsilateral excitatory pathway.

General response properties

Electrically elicited single-unit responses exhibited a number of properties that were qualitatively similar across all animal groups. Generally, fibers exhibited monotonically in-
Evidence of similar periods of suppression have been reported suggesting that the increases in latency seen in LT-17 recorded from a normal-hearing animal of equivalent age to animal (Hardie and Shepherd 1999). Although we cannot rule evoked field and evoked potentials recorded from the same with significant increases in the latency of the electrically-tended to increase with stimulus current to periods

Suppression of spontaneous activity during electrical stimulation is consistent with results from experiments using electrical stimuli (Kuwada et al. 1984; Semple and Kitzes 1985), the small number of nonmonotonic cells observed in the present study is consistent with previous findings using electrical stimuli (Snyder et al. 1991, 1995). Because the proportion of nonmonotonic neurons did not vary significantly with duration of deafness, any difference between the previous acoustic studies and the present may be related either to the nature of the stimulus or reflect reduced levels of inhibition associated with the deafening process.

In general we observed an increase in both first spike latency and temporal jitter as a function of hearing loss. Furthermore greater increases in both parameters were seen in the long-term deaf animal, suggesting that the extent of these changes are a function of duration of deafness. Any increase in latency and temporal jitter is likely to be a result of several mechanisms including an increase in the refractory properties of ANFs (Shepherd and Javel 1997) and a reduction in the efficiency of synaptic transmission along the ascending auditory pathway (Kotak and Sanes 1997). Finally, it is not surprising to observe an association between temporal jitter and latency in polysynaptic pathways such as the one from the auditory nerve to the IC, as jitter is added at each synapse along the pathway (Walmsley et al. 1998).

The prolonged latencies associated with LT-17 are consistent with significant increases in the latency of the electrically-evoked field and evoked potentials recorded from the same animal (Hardie and Shepherd 1999). Although we cannot rule out the effects of age, we have shown previously that ABRs recorded from a normal-hearing animal of equivalent age to LT-17 exhibited latencies similar to those of C-1 and C-2, suggesting that the increases in latency seen in LT-17 were not merely age-related (Hardie and Shepherd 1999).

Suppression of spontaneous activity during electrical stimulation was evident in both control and deafened animals. The suppression period had a clear threshold, and its duration tended to increase with stimulus current to periods >100 ms. Although this suppression may have originated within the IC, evidence of similar periods of suppression have been reported at the level of the dorsal CN in response to electrical stimulation of the auditory nerve (O’Leary et al. 1994; Shofner and Young 1985), suggesting its origins may have been within the brain stem.

Dynamic range was generally greater in neonatally deafened compared with abruptly deafened control animals. In part, these increases are likely to have originated from the auditory nerve, as increases in dynamic range have been observed in ANFs of long-term deafened animals (Shepherd and Javel 1997). Increases in the dynamic range of myelinated nerve fibers have been attributed to prolonged refractory periods and an increase in the vulnerability of the propagating spike associated with a loss of myelin and partial neural degeneration (Koles and Rasminsky 1972). It is also possible that there is a central component associated with a reduction in synaptic efficiency (Kotak and Sanes 1997).

For many units across all animals, a single pulse at near-saturation current level often elicited more than one excitatory spike. Because all animals in our series were deafened at the time of recording, this multiple spiking was not associated with electrophonic activation of hair cells. In deafened animals, multiple spiking in response to brief current pulses is not seen at the level of the auditory nerve (Shepherd and Javel 1997), although it has been observed in the CN (O’Leary et al. 1994) and the auditory cortex (Hartmann et al. 1997). The presence of multiple spiking within the central auditory pathway may reflect the convergent nature of afferent pathways terminating within these structures, recurrent neuronal circuits, or a reduction in the level of inhibitory processes within the ICC of deafened animals.

Given the precautions we previously have outlined associated with the interpretation of threshold, it is worth noting that in almost all cases in the present study, ipsilateral stimulation resulted in higher thresholds than contralateral stimulation. This observation is consistent with results from experiments using acoustic stimulation (Kuwada et al. 1984; Semple and

<table>
<thead>
<tr>
<th>Status</th>
<th>Median</th>
<th>Q₁*</th>
<th>Q₃</th>
<th>n</th>
<th>P†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0.235</td>
<td>0.180</td>
<td>0.430</td>
<td>30</td>
<td></td>
</tr>
<tr>
<td>Ipsilateral</td>
<td>0.351</td>
<td>0.250</td>
<td>0.430</td>
<td>30</td>
<td>0.246 NS</td>
</tr>
<tr>
<td>Bilateral deaf</td>
<td>0.380</td>
<td>0.185</td>
<td>0.558</td>
<td>39</td>
<td>0.430 NS</td>
</tr>
<tr>
<td>Unilateral deaf† +</td>
<td>0.540</td>
<td>0.428</td>
<td>0.738</td>
<td>5</td>
<td>0.022</td>
</tr>
</tbody>
</table>

All data were collected at saturation and are expressed in milliseconds. * Q₁, first quartile; Q₃, third quartile. † Mann Whitney rank sum test. Data compared with the appropriate side of the controls. †† Unilaterally deaf animals were deafened on the side contralateral to the IC recording site.

The scatterplot showing temporal jitter as a function of spike latency for 170 lateral IC (LN) and ICC neurons recorded from both the control and the neonatally deafened animals. All data were derived from stimulation of the cochlea contralateral to the recording site (which was the deafened cochlea in the case of the unilaterally deaf animals). Temporal jitter was measured at the lowest current level capable of evoking saturation ($P \geq 0.9$). There was no significant difference in either the slope or intercept of regression lines for each of the 3 groups of animals ($P > 0.05$; t-test), hence a single regression line derived from all the data are illustrated here. Equation for the fitted linear regression line together with its $r^2$ value are illustrated.
Kitzes 1985), and presumably reflects the dominant contralateral projection to the IC (Oliver 1987). In addition, the mean thresholds to both ipsilateral and contralateral stimulation in the long-term deafened animal were typically 2–3 dB greater than the control and neonatally deafened animals. This threshold increase is consistent with the elevated EABR thresholds recorded from the same animal (Hardie and Shepherd 1999) and probably is related to the significant loss of spiral ganglion cells and a significant increase in the distance between residual spiral ganglion cells and the stimulating electrode as a result of the deafness-induced degeneration of peripheral processes.

**LN versus ICC single-unit responses**

With the exception of the long-term deaf animal, electrical stimulation of the auditory nerve readily evoked excitatory activity from single units within the LN. Single units isolated from the LN in the present study displayed excitatory responses with latencies of ≤25 ms. As the recording electrode advanced toward the ICC, unit latencies tended to decrease to levels as low as 10 ms. In contrast, units isolated from the ICC exhibited latencies <10 ms and typically showed little variation throughout the track. Previous studies using electrical stimuli also have reported longer latency single-unit activity within the superficial region of the IC in contrast with shorter latencies found in the central nucleus (Shirane and Harrison 1991; Snyder et al. 1991), reflecting the known afferent auditory projections to this nucleus that appear to be dominated by third and higher order neurons (Kudo and Nakamura 1988).

**Response properties as a function of hearing status**

Although the long-term deaf animal exhibited extensive cochlear pathology, electrical stimulation of residual ANFs readily evoked single-unit activity within the ICC; however; the number of units isolated per millimeter of recording track...
was less in this animal than in the acutely deafened controls. Furthermore single-unit activity was evoked readily from animals deafened neonatally, a finding consistent with Snyder and his colleagues (1990, 1991, 1995). Our results contrast with those of Shirane and Harrison (1991), who could not reliably record excitatory activity from the IC of the neonatally deafened chinchilla. These authors concluded that a deafness-induced reduction in afferent activity in the auditory pathway during development results in an inactivation of IC neurons. It is difficult to reconcile the findings of Shirane and Harrison (1991) with our own and those of Snyder and colleagues. Our work, using neonatally and congenitally deaf animals, has shown no evidence of widespread inactivation at the level of the auditory nerve, auditory brain stem or auditory cortex (Hardie and Shepherd 1999; Hartmann et al. 1997; Shepherd and Javel 1997). As we have observed in the present study, a significant loss of ANFs associated with a hearing loss can result in elevated thresholds. It is possible that the stimulus levels required to evoke single-unit activity in Shirane and Harrison’s deafened chinchilla were outside the range of the search stimuli used in their study.

Why did we fail to isolate excitatory activity within the LN of the long-term deafened animal? Although a complete descrip-

FIG. 16. Input-output functions for 2 ICC neurons, illustrating the variability in following high-stimulus rates at high levels of entrainment. Neuron from the control animal (top) could be driven to saturation over a relatively narrow dynamic range (≈2 dB) for rates of ≈120 pps. In contrast, the neuron from the bilaterally deaf animal (bottom) showed a large increase in dynamic range as stimulus rate increased from 10 to 40 pps. At higher rates, the neuron failed to reach saturation. Both examples are in response to stimulation of the contralateral cochlea and illustrate near maximum levels of temporal resolution for the control and bilaterally deaf groups.

FIG. 17. Temporal response properties of ICC neurons from control and deafened animals, measured as the maximum stimulus rate for which each neuron could reach saturation (P > 0.9). Shown are the 5th, 25th, median, 75th, and 95th percentile for each group. LT-17 was not included in this analysis. Results based on n = 40 units from controls, n = 14 bilateral, n = 13 unilaterally deaf, n = 9 unilaterally control. * P < 0.05; Mann Whitney rank sum test.

IC response latencies

There appears to be little difference in the response latency of an IC unit to electrical stimulation from two distinct sectors of auditory nerve within the contralateral cochlea. These results were consistent with those of Snyder et al. (1991) and suggest that where ANFs from adjacent sectors project to the same neuron in the contralateral IC, they do so via a similar pathway.

FIG. 18. Scatterplot illustrating the relationship between 1st-spike latency and maximum stimulus rate for which each unit can reach saturation entrainment. There was no significant difference in either the slope or intercept of regression lines for contralateral vs. ipsilateral stimulation (P > 0.05; r-test), hence a single regression line derived from all the data are illustrated here. Equation for the fitted linear regression line together with its r² value are illustrated.
Our finding of a 7.5% shorter first spike latency for IC units to ipsilateral compared with contralateral stimulation in control animals was statistically significant and was consistent with the latencies of IC field potentials recorded from these animals (Hardie and Shepherd 1999). These results contrast with studies using acoustic stimulation where the response evoked via the contralateral ear is shorter than that evoked via ipsilateral stimulation (Semple and Kitzes 1985; review: Aitkin 1985)—consistent with the dominant contralateral projections to the IC.

The shorter latencies of ipsilaterally evoked responses in the present study suggest that excitatory activity is being relayed to the IC along the comparatively small direct projection from the ipsilateral CN rather than being relayed through the superior olivary complex and/or the lateral lemniscus. However, it is difficult to separate the effects of the nature of the stimulus from the effects of deafening. It may be that the highly synchronous nature of the electrical stimulus used in the present study was effective in evoking activity via the ipsilateral CN-ICC pathway; alternatively, a deafness-related reduction in inhibitory influences (Bledsoe et al. 1995) may have resulted in more effective activation of this pathway. Consistent with studies using acoustic stimulation, Vischer et al. (1997) reported longer ipsilateral compared with contralateral latencies of single units in the ICC of normal hearing adult rats evoked by electrical stimulation of the auditory nerve. These findings suggest that the reversal of the latency trend observed in the present study is not attributable to the nature of the electrical stimulus but rather relates to the deafening procedure and to its effects on the balance of excitatory and inhibitory influences on neurons within the ICC.

Cochleotopic organization of the ICC

A number of studies in normal-hearing animals have reported a systematic variation in threshold as a function of ICC recording depth in response to electrical stimulation of restricted sectors of auditory nerve (Black and Clark 1980; Merzenich and White 1977; Snyder et al. 1990). These studies have shown that single or multiunit thresholds produce a characteristic V-shaped STC as a function of recording depth, reaching a minima corresponding to the cochleotopic organization of the IC relative to the site of stimulation within the cochlea; stimulation of more apical sites within the cochlea produce threshold minima located more dorsolaterally within the IC compared with stimulation of more basal sites.

The present results extend these earlier findings, demonstrating the presence of a cochleotopic organization within the ICC of both control and neonatally deafened animals. Furthermore the presence of spatial tuning within the ICC of the long-term deafened animal suggests maintenance of at least a rudimentary cochleotopic organization within the IC in the absence of afferent input over many years.

Evidence of cochleotopic organization in animals deafened during the onset of normal auditory function is of particular interest. This finding is consistent with previous studies in both neonatally and congenitally deafened animals at the level of the auditory midbrain (Snyder et al. 1990) and cortex (Hartmann et al. 1997) and implies that cochleotopic organization within the auditory pathway is laid down—at least in a basic form—in the absence of normal afferent input during development. However, in the neonatally deafened animals, there is a decreased separation between the best locations for STCs generated by stimulation of the apical and basal electrode pairs compared with control animals. This may reflect changes at the level of the cochlea. In the neonatally deafened animals, spiral ganglion cell density was reduced to ~15% of normal, particularly in the upper basal and middle turns (Hardie and Shepherd 1999). A reduction in the number of neural elements within the periphery may result in a reduction in the spatial separation of the populations of neurons being stimulated by the two electrode pairs.

From the present study it appears that at least a rudimentary cochleotopic pathway is established at the level of the auditory midbrain before onset of normal auditory function and is therefore independent of sensory experience. This finding is consistent with other studies showing that gross connections of the auditory system are in place in altricial animals well before the onset of hearing (Friauf and Kandler 1990; Young and Rubel 1986). Although it is clear that excitatory activity is essential for normal neural development (e.g., Kalil 1990), the synaptic plasticity that shapes the mature pathway takes place late in development, refining more basic connections laid down under genetic cues. This modification appears to depend initially on spontaneous activity (Miller 1994)—ANFs are known to be spontaneously active before 10 DAB (Romand 1984; Walsh and McGee 1987)—and a later period of development during which established networks are modified by experience (Feldman and Knudsen 1998).

Temporal resolution

The range of electrical pulse rates to which IC units responded in a temporally synchronized manner showed wide variation but were typically < 120 pps. In addition, there was a significant negative correlation between first spike latency and temporal resolution. Similar observations have been reported by others using both acoustic (Langner and Schreiner 1988) and electrical (Snyder et al. 1991, 1995) stimuli.

A significant finding in the present study was evidence of reduced temporal resolution of neurons isolated from neonatal bilaterally deafened animals compared with controls. In contrast, neurons from neonatal unilaterally deafened animals showed no evidence of such a reduction. Presumably monaural input is sufficient to maintain neurons within the auditory midbrain in a state in which they exhibit normal temporal resolution.

Although there were differences in the measure of temporal resolution between the two studies, Snyder and colleagues also observed a reduction in the temporal resolution of cells in bilaterally deafened animals albeit not statistically significant (Snyder et al. 1995). They did, however, report a significant increase in the temporal resolution of IC neurons in bilaterally deafened animals that had received afferent input via chronic electrical stimulation of the auditory nerve (Snyder et al. 1995). Together, these results indicate the importance of afferent input in maintaining—or enhancing—the temporal resolution of IC neurons.

Previous studies using acoustic stimuli have demonstrated the generally low temporal resolution of IC units compared with ANFs or cells within the CN (Kiang et al. 1965; Langner and Schreiner 1988; Moller 1974; for review, see Irvine 1986). Data obtained via electrical stimulation of the auditory nerve...
are in agreement with the acoustic data. Although the great majority of neurons within the IC fail to respond at high entrainment levels to current pulses >120 pps, ANFs readily respond at high levels of entrainment to current pulses presented at 800–1,000 pps (Hartmann et al. 1984; Javel et al. 1987; Moxon 1971; Shepherd and Javel 1997; van den Honert and Stypulkowski 1984). Neurons in the primary auditory cortex show further reductions in temporal resolution compared with cells in the auditory midbrain (Schreiner and Reggio 1996). These reductions in temporal resolution presumably are related to a systematic increase in the number of synapses along the ascending auditory pathway for progressively higher centers.

Clinical experience with cochlear implants in congenitally deaf adults and children

Adult postlinguistically deafened implant recipients show a wide range of speech perception skills. Factors enhancing speech perception ability include auditory experience before implantation and with implant use, whereas duration of deafness shows a strong negative correlation with speech perception (Blamey et al. 1996). A number of studies have shown that adult prelinguistically deafened implant patients show poor speech perception compared with adults deafened after language development. Psychophysical studies have shown that prelinguistically deafened adults generally have electrode pitch percepts that show no consistent variation with tonotopic organization (Busby et al. 1992; Eddington et al. 1978). These results are consistent with the observed reduction in separation between the spatial tuning curves of the neonatally deafened animals in this study. More recent evidence using younger subjects indicates that ~60% of subjects show a clear and consistent tonotopic order of pitch percepts, with poorer performance associated with longer periods of auditory deprivation (P. A. Busby, personal communication).

The temporal processing skills of prelinguistically deafened implant patients have been shown consistently to be poor compared with postlinguistically deafened patients (Busby et al. 1992; Eddington et al. 1978). These subjects show a reduced ability to perceive different rates of stimulation and reduced gap detection abilities. It is of interest to note that there is evidence of an improvement in these temporal processing skills with device use, a finding that is consistent with evidence of increased temporal resolution of ICC neurons in neonatally deafened animals after chronic electrical stimulation of the auditory nerve (Snyder et al. 1995).

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REFERENCES


