Cortical Representation of Interaural Time Difference in Congenital Deafness

Binaural cues are required for localization of sound sources. In the present paper, representation of binaural cues has been investigated in the adult auditory cortex. Hearing and congenitally deaf cats were stimulated through binaural cochlear implants and unit responses were collected in the subregion of field A1 showing the largest amplitudes of evoked local field potentials. Sensitivity to interaural time difference (ITD) in the range from $-600$ to $600\,\mu$s was tested at intensities of 0–10 dB above hearing threshold. Template ITD functions were fitted to the data and parameters of ITD functions were compared between deaf and hearing animals. In deaf animals, fewer units responded to binaural stimulation, and those that responded had smaller maximal evoked firing rate. The fit to the template ITD functions was significantly worse in deaf animals, and the modulation depth in ITD functions was smaller, demonstrating a decrease in ITD sensitivity. With increasing binaural levels, hearing controls demonstrated systematic changes in ITD functions not found in deaf animals. Bimodal responses, likely related to precedence effect, were rare in deaf animals. The data demonstrate that despite some rudimentary sensitivity to interaural timing, cortical representation of ITDs is substantially altered by congenital auditory deprivation.

Keywords: auditory deprivation, binaural, cochlear implant, development, spatial hearing

Introduction

Some features of cortical processing develop very early after hearing onset, whereas others reach maturity only after a period of hearing experience (cat: Eggermont 1996; Bonham et al. 2004). Elementary binaural interactions can be demonstrated at postnatal day 8 in the cat, before the hearing thresholds have declined below 100 dB sound pressure level (SPL) (Brugge et al. 1988). Binaural information (interaural time and level differences) as well as spectral shaping of sounds by the pinna are used for sound-source localization. Interaural intensity difference is the dominant cue for localization of high-frequency sounds, whereas interaural time difference (ITD) for localization of low-frequency sounds. ITD is also involved in localization of high-frequency and complex sounds using the signal envelope. Sensitivity to ITDs is established in the superior olive and depends on the temporal coincidence of binaural inputs as well as on additional factors like cochlear disparities (Joris et al. 2006; Joris and Yin 2007) and inhibition (Brand et al. 2002; Grothe 2003; McAlpine and Grothe 2003; McAlpine 2005). The development of inhibition is affected by continuous presentation of noise that disrupts the ITD information (Kapfer et al. 2002; Seidl and Grothe 2005).

Despite decoding of interaural information at the level of the brainstem, the behavioral response to sound location requires the primary auditory cortex. Lesion or reversible inactivation of field A1 leads to impairment in auditory localization performance (Neff 1977; Jenkins and Merzenich 1984; Malhotra et al. 2004). In humans, case reports demonstrate that lesions in the primary auditory cortex lead to sound localization deficits (Clarke et al. 2002; Adriani et al. 2003). Thus, activity in the primary auditory cortex is an essential (but not sufficient) condition for sound-localization performance.

The auditory system shows ITD sensitivity in the range of microseconds. To investigate whether this sensitivity is preserved even in the absence of hearing experience, congenitally deaf cats (CDCs) were used as a model of congenital auditory deprivation. They were identified from a colony of “deaf” white cats by a hearing screening procedure early in life (Heid et al. 1998). CDCs are known to have a functional afferent auditory pathway (Hartmann et al. 1997; Heid et al. 1997). The spiral ganglion degenerates very slowly, over years (Heid et al. 1998). In terms of spiral ganglion degeneration, CDCs compare much better with human prelingual deafness (Felix and Hoffmann 1985; Pollak and Felix 1985) than other deafness models. Comparisons between various brain structures have not revealed any nonauditory deficits (West and Harrison 1973; Saada et al. 1996; Kral et al. 2003; Lomber et al. 2008). On the other hand, numerous auditory deficits have been demonstrated in CDCs, including reduced cortical synaptic activity and changes in microcircuitry of the cortical column (Kral et al. 2000) caused by a delayed and modified postnatal development (Kral et al. 2005). These and other deficits lead to a sensitive period in cortical plasticity for restoration of hearing with cochlear implants (CIs) (Kral et al. 2002, 2006).

With regard to binaural hearing, cochlear-implanted people have difficulties with differentiation of ITD (van Hoesel and Tyler 2003; Senn et al. 2005; Seeger and Fastl 2008). Clinical use of binaural CIs makes information on central binaural processing a clinically relevant topic (Van Deun et al. 2009). The present study aims to demonstrate that sensitivity to ITD is preserved in a few cortical units even in congenital deafness, yet its cortical representation is significantly compromised.

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Materials and Methods

The present study was carried out on 8 animals: 4 congenitally deaf and 4 hearing cats, all at adult age (>12 months). CDCs are deprived from auditory experience: They lose the hair-cell function between postnatal days 3 and 10 (Mair and Elverland 1977), before hearing thresholds decline under 100 dB SPL in normal cats. They do not develop brainstem-evoked responses (Mair and Elverland 1977). The deafness is spontaneous, inherited and nonprogressive (Heid et al. 1998). For the present study, CDCs were selected from a colony of deaf white cats by the absence of brainstem-evoked responses to click stimulation at >120 dB SPL in the fourth postnatal week (Heid et al. 1998). Total deafness of the animals was additionally confirmed at the beginning of the acute experiments with the same procedure. To prevent electrically hearing (electrical stimulation of hair cells) in hearing controls (HCs), these animals were deafened at the beginning of the experiment by intracochlear application of neomycin (Hartmann et al. 1984). In what follows, HCs refer to normal hearing animals acutely deafened at the time of the experiment. The adjective "hearing" does not refer to the functional state of the cochlea during the experiment but to the developmental and functional state of the central auditory system (hearing experience). The experiments were approved by the local state authority and were performed in compliance with the guidelines of the European Community for the care and use of laboratory animals (EU VD 86/609/EEC) and the German law for protection of animals.

Cochlear Implant

The CI was a medical-grade silicone tube with 5 intrascalar contacts: a small golden ball at the tip (diameter 0.8 mm) and 4 golden rings, with a distance of 1 mm between the electrode contacts (Behrendt 1999). The intrascalar part of the implant was tapered in the apical direction from a diameter of 1.6 to 0.8 mm. The gold contacts were connected to a 7-strand Teflon-coated stainless-steel braided wire. The stimulation mode was wide bipolar (most apical vs. the fourth intracochlear electrode).

Experimental Procedures

All animals were premedicated with 0.25 mg atropine i.p. and initially anesthetized with ketamin hydrochloride (24.5 mg/kg Ketavet, Parker- Davis, Germany) and propionylpromazine phosphate (2.1 mg/kg Combelen, Bayer, Germany). The animals were then tracheotomized and artificially resipilated with 50% O2 and 50% N2O, with the addition of 0.2-1.5% concentration of isoflurane (Lilly, Germany) to maintain a controlled depth of anesthesia (Kral et al. 1999). End-tidal CO2 was monitored and maintained below 4%, and core temperature was kept above 37.5 °C using a homeothermic blanket. The animal’s status was further controlled by blood gas concentration measurements, pH, bicarbonate concentration and base excess, and glycemia and oxygen saturation. A modified Ringer’s solution containing bicarbonate and plasma expander was infused i.v. with additional bicarbonate depending on the acid-base status. Monitoring and correction of acid-base were performed every 12 h.

The animal’s head was fixed in a stereotactic holder (Horsley-Clarke). Both bullae and ear canals were exposed. In order to record evoked auditory brainstem responses, a small trephination was drilled at the vertex and a silver-ball electrode (diameter 1 mm) was attached epidurally. The indifferent electrode used for the recordings was inserted medially in the neck muscles.

Hearing status was verified using brainstem-evoked responses with condensation clicks (50-μs duration) applied through a modified DT 48 speaker (Bayer Dynamics, Germany). The speaker membrane was positioned ~1 cm from the tympanic membrane within a custom-made acoustic calibrator. Preamplifying the calibrated sound delivery device (closed system) inserted into the remaining part of the external auditory meatus after the pinna was removed. Brainstem-evoked signals were recorded using an epiperal vertex electrode against a reference at the midline of the neck, were preamplified (60 dB, Otoconsult V2 Low-impedance Amplifier), amplified in a second stage (40 dB, Otoconsult Amplifier-Filter F1, filters 0.010-10 kHz), and recorded using National Instruments MIO Cards and averaged (200 sweeps, repetition rate 33 Hz, AudiologyLab, Otoconsult Comp., Frankfur am Main). The absence of acoustically evoked brainstem responses (EBR; including wave I, generated within the auditory nerve) to clicks greater than 120 dB SPL verified complete deafness. In HCs, the thresholds were less than 40-dB SPL before the animals were deafened by a slow instillation of 300 μL of neomycin sulfate into the scala tympani (within 5 min). The neomycin was left in place for further 5 min and was subsequently washed out by slow instillation of Ringer’s solution. Absence of brainstem-evoked responses verified a successful deafening procedure.

For electrical stimulation, the animals were bilaterally implanted with a CI electrode array inserted via the round window in each cochlea. Comparable insertion depth between the 2 ears was controlled by inserting the electrode array until the most basal electrode of the implant was just observed behind the rim of the round window (insertion depth ~6 mm). The bulla was sealed with bone wax. Charge-balanced pulses (200 μs/phase, repetition rate 2 Hz) were applied to the CI (wide bipolar stimulation)—most apical vs. fourth electrode, interelectrode distance ~4 mm). Stimulation was performed with optically isolated current sources (CS1, Otoconsult, Germany). To verify comparable position of the CIs, electrically EBR with single biphasic pulses were recorded and lowest current levels evoking a brainstem response ("EBR threshold currents") were determined by visual inspection. If the thresholds for stimulation at the left and the right ears differed by more than 2 dB, the ear with higher thresholds was reimplanted and the thresholds were remeasured.

Trephination was performed above the auditory cortex and the dura was removed. The cortex was photographed to document the recording positions. Using an x-y-z micromotor (1 μm precision in all directions), a silver-ball macroelectrode (diameter 1 mm) was positioned at 9 cortical positions on the primary auditory cortex (field A1). The dorsal end of the posterior ectosylvian sulcus was used as a reference point. Signals were preamplified (60 dB, Otoconsult V2 low-impedance amplifier), amplified at a second stage (20 dB, Otoconsult F1, filters 0.010-10 kHz), recorded using NI MIO Cards and averaged (100 sweeps, repetition rate 1.97 Hz, AudiologyLab, Otoconsult Comp., Frankfur am Main). The signals were stored and "cortical threshold current levels" were determined at all recording positions with a precision of ±1 μA. These were determined for cortical mapping purposes only.

In order to determine the extent of the cortical activated region, a Ringer-filled “glass microelectrode” (impedance <6 MΩ) was used. Local field potentials (LFPs) on the cortical surface were recorded at 75-150 cortical positions during stimulation with the CI using biphasic pulses (200 μs/phase, wide bipolar stimulation at the ipsilateral and also contralateral ear, stimulation current 10 dB above the lowest cortical threshold determined with the macroelectrode). Fifty to 100 responses were averaged to obtain evoked LFPs. Amplitudes of middle-latency responses (peak-to-trough) were used to construct cortical activation maps using a custom-made software programed in MatLab (MathWorks). Convergence of bilateral activity in the same locations of the primary auditory cortex was verified in all investigated animals.

After determining the functional activation map in the cortex, the cortical tissue was penetrated at the cortical spots with the largest LFPs ("hot spots") using a "Michigan Probe" (NeuroNexus, United States, 16 contacts, spacing 150 μm, 40 x 40-μm contacts, impedance ~1-2 MΩ) such that the last electrode was placed at the level of the cortical surface. The movement of the cortex was stabilized using a modified Davies chambers filled with agar and sealed with melted bone wax.

The signals were amplified by a 32-channel Cheetah amplifier (Neuralynx, Tuscon, AZ, ×2000-5000 amplified, Filters 1 Hz-9 kHz), fed to the input of a National Instruments MIO Card, and stored on a computer using custom-made MatLab routines. Filtering and unit selection were performed off-line. The unit recording was focused on the hot spot determined within the cortical map. Figure 1 shows all recording positions with the Michigan probe from all animals pooled with the posterior and anterior ectosylvian sulcus used as a reference.

Binaural Stimulation

Binaural stimulation was performed using a train of 3 biphasic charge-balanced pulses (200 μs/phase) with a repetition rate of 500 pps. Thus, the total stimulation duration was <5 ms in all binaural conditions.
Thirty to 50 responses to stimulation of each ear were recorded separately with increasing intensity (1- to 2-dB precision, interstimulus interval of 623 ms). Afterward, with reference to the individual EBR thresholds, intensities at both ears were covaried. EBR threshold was used as a reference so as to compensate for the possible differences in the exact location of the CIs within the scala tympani. ITDs were varied in the range of 0-600 μs (0, 10, 20, 50, 100, 200, 300, 400, 500, and 600 μs), with stimuli leading at both ipsilateral and contralateral ears. Each stimulus condition was repeated 30-50 times so that robust response properties could be determined.

Stimulation was controlled through visual inspection of current monitor signals for both current sources (at contralateral and ipsilateral ears) on an oscilloscope throughout the whole experiment.

Data Processing
Recorded signals were processed using custom-made routines programed in Matlab (Mathworks). Data were high-pass filtered (elliptic filter, second-order, cut-off frequency of 300 Hz), and stimulus artifacts were blanked. Zero-phase digital filtering was performed to avoid action potential latency shifts. Unit activity was detected by an automatic thresholding procedure (Quiroga et al. 2004, for details see Supplementary Material).

Multi-units were considered to respond to the stimuli if the spike pattern in analyzed poststimulus time windows could not be explained by a Poisson process using the spontaneous rate observed in the prestimulus time window (Chase and Young 2007; for details, see Supplementary Material). Two poststimulus time windows were analyzed (0-15, 15-50 ms, respectively, Fig. 2).

To compare controls with deaf cats, ITD functions from the first response (poststimulus time 0-15 ms) were automatically classified at all stimulus intensities using custom-made software. Templates for classification were similar to a previous study (Smith and Delgutte 2007; for technical details and the equations see Supplementary Material). Four different functions were defined: peak, trough, sigmoid, and biphasic (Fig. 7a). In each of these functions, 4 parameters were evaluated: ITDbest is the parameter defining the ITD with the strongest (peak, sigmoid, and biphasic function) or weakest (trough function) response. As the name suggests, this parameter possibly quantifies the interaural time represented by the given unit. ITDcenter was defined as the point in the center of the rising phase of the template function (cf. Fig. 7a). This parameter represents the point of the highest contrast sensitivity, as at this point of the function changing ITD evokes the highest change in firing rate. To characterize the "ITD tuning," half-rise (HR) represents the half-width of the rising phase of the function, corresponding to the half-width of the template in its half amplitude (Fig. 7a). Finally, modulation depth (MD) of the responses was defined as the ratio of the maximal and minimal response firing rates. That parameter thus characterizes the dynamic range of the ITD function.

Responsive ITD functions with nonsignificant fits to either template (determined by the correlation between data and fitted function; significance level for 18 degrees of freedom at $r < 0.56$, $\alpha = 1\%$) were considered "nonsignaled" ITD responses. ITD functions with MD less than 50% were classified as "flat." All parameters obtained from the fitted templates (flat class excluded) were compared between HCs and CDCs at 0, 3, 6, and 9 dB above the individual EBR threshold.

Complementarity of the ITD function computed from the first (0-15 ms) and the second (15-50 ms) response (Fig. 2) was evaluated by correlating ITD functions in the 2 analyzed time windows. Instead of the
Figure 3. Normalized population rate–intensity functions relative to individual unit threshold (a–d) and EBR threshold (e–h) in HCs (left) and CDCs (right). Shown are mean firing rates normalized to maximum (black line) and 1 standard deviation (shaded area) for all responsive units. Deaf animals demonstrate changes in representation of stimulus intensity.
templates, here original data were correlated. Only ITD functions with MDs greater than 50% were tested for complementarity. If the first and the second response ITD functions were negatively correlated with a correlation coefficient less than \(-0.5\), the recording site was considered to have complementary responses in the 2 analyzed time windows.

For quantification of inhibitory effects within ITD functions, ITD\textsubscript{contrast} was introduced:

\[ \text{ITD}_{\text{contrast}} = \frac{\text{ITD}_{\text{max}} - \text{ITD}_{\text{min}}}{\text{ITD}_{\text{max}} - \text{SR}}; \]

whereas ITD\textsubscript{max} corresponds to the firing rate observed at the maximum of the ITD function, ITD\textsubscript{min} corresponds to the firing rate observed at the minimum of the ITD function and SR corresponds to the spontaneous activity at the given site in the absence of the stimulation (Nishino et al. 2008).

Results were statistically compared by nonparametric 2-tailed Wilcoxon-Mann-Whitney (WMW) test or Kolmogorov-Smirnov (KS) test.

Results

Using cortical mapping with surface-recorded LFPs, 2-3 spots of high activity between the anterior and posterior ectosylvian sulcus were reproducibly observed (see Kral et al. 2009). The position of these hot spots was well reproducible in all investigated animals (ibid.). The hot spots for the ipsilateral and contralateral stimulations overlapped in all animals investigated (cf. ibid.).

Unit recordings were measured in field A1 within a region of interest defined by the largest LFPs (spots HS1 and HS3 in Kral et al. 2009). On average, 84 unit recordings were included into the analyses for each animal. Recording positions covered a similar cortical region in both groups of animals (Fig. 1).

Temporal Structure of the ITD Responses

To eliminate the stimulus artifact, 6 ms poststimulus were blanked from the recording. Multi-unit responses to binaural stimuli typically showed either unimodal or bimodal distribution in poststimulus time histograms (PSTHs) (Fig. 2). The first peak in the PSTH had a latency between 6 and 15 ms poststimulus, and the second peak had a latency between 15 and 50 ms.

Responses were obtained at several binaural intensities above individual unit thresholds. Mean maximal firing rate for contralateral stimulation was 1.03 spikes/stimulus in HCs and 0.44 in CDCs (WMW test, \( P \ll 0.0001 \)), and for ipsilateral stimulation 0.58 spikes/stimulus in HCs and 0.49 in CDCs (WMW test, \( P = 0.17 \)). The maximum firing rate was significantly higher for contralateral than ipsilateral stimulation in HCs (\( P \ll 0.0001 \)) but not in CDCs (\( P = 0.52 \)). Firing rates were further normalized with respect to the maximum firing rate and averaged at each stimulation intensity (Fig. 3a-d). In HCs, the resulting population rate-intensity functions for monaural stimulation showed a dynamic range of 4–6 dB (Fig. 3a,c). The dynamic range was reduced by deafness, with the steepest increase in population-firing rate within the first 2 dB above threshold (Fig. 3h,d).

However, in vivo, the cortex has no access to the firing rate of its units relative to the individual unit’s threshold; rather, the given stimulus evokes a spatiotemporal pattern that is related to the stimulus intensity relative to the hearing (cochlear) thresholds. Therefore, cortical units were also recomputed relative to EBR thresholds, which were taken as the measure of the cochlear hearing thresholds. In HCs, only few units showed evoked responses within the first 3 dB above threshold.
population rate–intensity function demonstrated a prominent increase in firing rate at 4–6 dB (Fig. 3b, g). In deaf animals, this systematic "arrangement" of unit thresholds to EBR threshold was not apparent and a more monotonic firing rate function with increasing intensity was observed.

The number of nonresponsive recording sites was significantly greater in CDCs than in HCs (deaf: 43.8 ± 9.8%, controls: 10.8 ± 8.3%, WMW test, P = 0.028).

**ITD Sensitivity**

The firing rate was dependent on ITD in HCs (Fig. 4). Within the same single-shank Michigan probe, similar and different ITD sensitivities were repeatedly observed. In CDCs, the dependence of firing rate on ITD was less systematic in the majority of units (Fig. 4). To be able to quantify the ITD sensitivity, "classical" ITD functions (e.g., Brugge and Merzenich 1973) were constructed from the first response (Fig. 5). In HCs, these ITD functions showed systematic changes in firing rate with ITD, which were well reproducible with increasing binaural stimulus intensity. Also, in CDCs, units with a systematic relation of firing rate to ITD were occasionally observed (Fig. 5), clearly demonstrating rudimentary ITD sensitivity in congenital deafness. In contrast to acoustic stimulation, these functions sometimes showed an additional local peak near 0 μs (Fig. 5, asterisks).

For each ITD function, the maximum firing rate was determined. Responsive units showed a significant and extensive reduction in the mean maximum evoked firing rate.

![Figure 5](https://academic.oup.com/cercor/article-abstract/20/2/492/312302)

Figure 5. Examples of ITD functions computed from the first response window at different binaural intensities for HCs (upper row) and congenitally deaf (below). Tuning to certain ITD values is obvious in all units shown in both groups of animals. Increasing the binaural intensity level above unit threshold (inset) did not change the ITD sensitivity. Asterisks indicate local peaks near 0 μs (for details, see text).
in CDCs (Fig. 6; controls: 1.62 spikes/stimulus; deaf: 0.72 spikes/stimulus; and WMW test, \( P < 0.001 \)). A decrease in maximum evoked firing rate was also observed in the second processing window (second response, Fig. 6).

To allow quantitative comparisons among the large number of ITD functions, an automatic classification was used. Starting from 6 dB, the population rate–intensity functions increased in HCs (Fig. 3e) such that the majority of units responded at this stimulation level. Therefore, comparisons were first performed at 6 dB above EBR threshold from the first response. Four different ITD template functions were used (Fig. 7).

Although the number of nonclassified units was not significantly different between deaf and hearing cats, the mean correlation of the templates with the data ("goodness of fit") was significantly lower in CDCs than in HCs (Fig. 7c; deaf: 0.82 ± 0.12; controls: 0.88 ± 0.11; WMW test, \( P = 0.000023 \)). Thus, the firing rate varied less systematically with ITD in CDCs than in HCs (cf. Fig. 4). This demonstrates a reduction of signal-to-noise ratio in ITD functions in deaf animals.

For the set of responsive recording sites, the proportion of different response types was determined (Fig. 7d). In HCs, the peak and sigmoid types were most frequent, whereas trough units were rare. There was no significant difference in the relative frequency of occurrence of individual response types between HCs and CDCs (Fig. 7d; WMW test, \( \alpha = 5\% \)).

The parameters of the fitted templates were consequently compared between hearing and deaf animals at 6 dB above threshold (Fig. 8a–d). Although visual inspection indicated a slight decrease in ITD_{best} in deaf animals, the difference was not statistically significant (WMW test, \( P = 0.58 \)). For the other parameters, only MD was significantly smaller in deaf animals (Fig. 8d; WMW test, \( P = 2.41 \times 10^{-4} \)). This demonstrates a decrease in the dynamic range of the ITD response and thus a reduction of ITD sensitivity in CDCs.

**Dependence on Stimulus Intensity**

Visual inspection of some ITD functions revealed remarkable stability with increasing binaural current levels in HCs (Fig. 5). If a change in the best ITD with increasing stimulus levels was observed, it was, in majority of cases, directed toward smaller ITDs. In deaf cats, some recorded units showed ITD functions with a similar stability with respect to increasing stimulus intensity as units in controls (Fig. 5).

At the population level, the membership in ITD classes changed with increasing binaural levels in HCs. The number of sigmoid responses decreased and the number of peak responses increased (Fig. 9a). A tendency for an increase in flat responses with increasing binaural levels was observed. In deaf animals, no such changes were found (Fig. 9b). Increasing binaural levels significantly decreased ITDbest in HCs (Fig. 9c for details, see Supplementary Material) but not in deaf animals (Fig. 9d). Correspondingly, the "center" of the ITD functions (the point of their highest contrast sensitivity) moved to smaller ITDs only in HCs (cf. Fig. 9e to Fig. 9f). At 3 dB above threshold, the ITD_{center} was significantly larger in controls than in CDCs, whereas at 9 dB, it was significantly smaller (see Supplementary Material). Consequently, with increasing binaural levels, the difference in ITD_{center} between HCs and CDCs reversed due to the systematic decrease of ITD_{center} in HCs.

ITD functions did not broaden with increasing intensities: There was no change in ITD_{half-rise} neither in HCs nor in CDCs (Fig. 9g,h). MD decreased with increasing binaural levels only in HCs (Fig. 9f). This was because of increasing responsiveness at the "worst" ITD and higher relative occurrence of flat responses. This was not observed in CDCs, who had a low MD already at the lowest intensities (Fig. 9f).

To further investigate the morphology of ITD functions, ITD_{contrast} was introduced, reflecting the difference in firing rate at worst and best ITDs with reference to the spontaneous activity (Fig. 10, Nishino et al. 2008). This index is particularly useful to indicate whether the firing rate rises at all ITDs above the level of spontaneous activity or whether some ITDs do not evoke any increase in firing rate and instead decrease the firing rate below the spontaneous level (Fig. 10b). The latter behavior indicates the action of neuronal inhibition. Such behavior was frequently observed in hearing cats (ITD_{contrast} > 1), but only occasionally in deaf cats (Fig. 10c). HCs showed a significant decrease in ITD_{contrast} with increasing binaural levels (WMW test, \( P = 0.04 \)) between 0 and 3 dB, \( P = 0.0012 \) between 3 and 6 dB, \( P = 0.096 \) between 6 and 9 dB), which was not found in CDCs (WMW test, \( P \) in the range from 0.20 to 0.91). There was a significant difference in ITD_{contrast} between deaf and control animals at lower binaural levels (WMW test; 0 dB: \( P < 0.001; \) 3 dB: \( P = 0.00092; \) 6 dB: \( P = 0.014; \) 9 dB: \( P = 0.136 \)).

Finally, ITD sensitivity was investigated as a function of response latency. All responses were classified into unimodal and bimodal categories, the latter showing significant responses in both analyzed time windows (0–15 and 15–50 ms). To include all responsive units, the analysis was performed at the highest stimulation intensity (9 dB above threshold). If bimodal responses were encountered, ITD functions of the first and second responses were compared. In bimodal units of HCs, ITD functions had highest and lowest firing rates in the 2 response windows at either complementary ITDs (where the first response had a maximum rate, the second had a minimum rate) or similar ITDs (Fig. 11c,d and e,f, respectively). To be considered complementary, the ITD functions had to be negatively correlated (\( r < -0.5 \), see Materials and Methods). Most frequently, responses in both time windows were observed in HCs, whereas these were primarily noncomplementary (Fig. 11g). In CDCs, responses

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**Figure 6.** Evoked firing rates obtained from the maximum of the ITD functions in both groups of animals and both response windows. CDCs achieved significantly lower maximum firing rates than HCs in both response windows (2-tailed WMW test, \( P < 0.001 \)). For details see text.
Bimodal responses were rare in deaf cats, both those with complementary ITD tuning (controls: 13.7 ± 9.4% of all recording sites; CDCs: 2.7 ± 2.3%; WMW test, P = 0.028), and those with noncomplementary ITD tuning (controls: 58.7 ± 14.8% of all recording sites; CDCs: 25.0 ± 11.5%; WMW test, P = 0.028).

**Comparison of the Caudal and Rostral Activation Spots**

As best ITD is a function of best frequency (McAlpine et al. 2001), all statistics were also performed in each hot spot separately at 6 dB above threshold. Although these location-specific data were consistent with the above findings, some differences were observed between these spots in HCs. ITD<sub>best</sub> was smaller in the rostral spot (caudal spot: 253.6 ± 367.2 μs, rostral spot: 155.9 ± 327.7 μs, 2-sample KS test P = 0.000562). This difference was not observed in deaf animals (caudal spot: 111.1 ± 352.0 μs, rostral spot: 350.2 ± 524.9 μs; KS test, P = 0.201). Comparison of the ITD<sub>center</sub> showed a similar result (controls: caudal spot: −168 ± 231.6 μs, rostral spot: −121.4 ± 234.5 μs, KS test P = 0.000305; deaf: caudal spot: −105.9 ± 290.8; rostral spot: 69.55 ± 300.4; KS test, P = 0.306). In consequence, in HCs, the contralaterality was significantly
greater in the caudal spot (as revealed by medians of ITD_best for caudal/rostral spot: 227.7/109.3 μs). In CDCs, this difference disappeared (135.9/113.3 μs). These results demonstrate further changes in cortical ITD representation in CDCs.

ITD functions fit significantly better in caudal than in rostral spot in HCs (caudal spot: 0.89 ± 0.11, rostral spot: 0.86 ± 0.09, WMW test, \( P = 0.034 \)). This difference was also not observed in CDCs (caudal spot: 0.83 ± 0.12, rostral spot: 0.82 ± 0.13, WMW test, \( P = 0.859 \)). No significant differences were found between the spots in half-rise and in response classes in both groups of animals.

**Discussion**

The present paper demonstrates for the first time profound deficits in the representation of ITD in deaf animals. In CDCs, fewer cortical units responded to binaural stimulation, and those that did respond showed a reduced maximum evoked firing rate. Additionally, the fit to the ITD templates was weaker in deaf animals and the MD was reduced, demonstrating a weaker sensitivity to ITDs. ITD functions were intensity dependent in HCs, which was not the case in deaf cats. Consequently, cortical representation of ITD requires auditory experience.

However, some residual sensitivity to ITD was observed in CDCs. Because cortical units have binaural properties inherited from the subcortical stages of processing (Nelken et al. 2008), the present data imply that, although cortical representation of binaural information requires auditory experience (see also Kral et al. 2009), some subcortical analysis of ITD is preserved in congenital deafness.

**Methodological Considerations**

The position of the electrode array within the scala tympani influences the hearing thresholds (Shepherd et al. 1993). To compensate this effect, current levels were given relative to EBR thresholds in the present study.

ITD sensitivity was tested for a set of time differences that well exceeds the physiological range in the cat (± 400 μs). Therefore, peaks in the ITD functions should be well represented in the sample of recordings performed in this study. The peak in ITD sensitivity was well within the physiological range. Only sigmoid functions did not have a clear peak in the investigated range, some of them even at the highest intensity tested. This indicates that they belong to neurons with excitatory–inhibitory class of responses inherited from the lateral olivary complex (Lohuis and Fuzessery 2000). Comparisons of ITD sensitivity and monaural response properties will be the focus of a follow-up study.

Due to the short duration of the auditory stimuli (5 ms, 3 stimuli), the present ITD functions reflect onset ITD sensitivity (Hirsch et al. 1985; Yin et al. 1985) rather than ongoing ITD sensitivity (Joris et al. 2008). Onset ITD sensitivity is of cardinal importance for orientation in space, initiating head and pinna orientation reflexes.

Here, multi-unit activity yielded comparable ITD sensitivity for HCs as those obtained with single units (inferior colliculus: Kuwada and Yin 1983; McAlpine et al. 2001; Smith and Delgutte 2007; auditory cortex: Reale and Brugge 1990). However, from the maximum firing rates of multi-units, conclusions cannot be drawn on the firing rates in single cells. Some nonclassified ITD functions might arise from recording a mixture of single units, although such functions have been also found with single units (Smith and Delgutte 2007).

**Classification of ITD Functions**

The present study used an automatic classification of ITD functions based on the scheme proposed by Smith and Delgutte (2007). However, the definition of the ITD classes differs among different authors (for intermediate, peak, and trough classes, see Fitzpatrick et al. 2000; Coffey et al. 2006). The main goal of the present study was the comparison...
between deaf and hearing animals. The results of the classification and statistical comparison of the best fitted template parameters allow a precise reconstruction of the position of peaks/troughs of the individual functions. The present data were also fit to other, more “flexible” templates (results not shown), yielding very similar results.

The present study performed using standardized multi-electrode arrays could for the first time automatically process

Figure 9. Changes of fitted template parameters with increasing binaural intensity (level), HCs on the left, CDCs on the right. (a,b) Statistics of the distribution of ITD classes in controls and deaf animals as a function of intensity (3, 6, and 9 dB levels shown from left to right). (c,d) ITDbest decreased with increasing binaural stimulus levels in HCs but not in deaf animals. (e,f) ITDcenter decreased with increasing binaural levels in HCs but not in deaf animals. (g,h) ITDwidth did not depend on binaural stimulus level in either group. (i,j) MD decreased with increasing binaural stimulus intensities in HCs but not in deaf animals.
all data as collected, without a bias toward "nicer" units, nicer ITD functions, or better responsive cortical layers. The technique also allowed us to assess the proportion of nonresponsive recording sites.

**Effect of Electrical Stimulation**

The distribution of ITD classes as well as the ITD parameters obtained in controls are similar to distributions obtained in a previous study using CI stimulation in acutely deafened cats (Smith and Delgutte 2007), where the mean ITD_{best} was 161 μs compared with 216 μs observed here. The present result is within the range obtained with acoustical stimulation (Brugge and Merzenich 1973; Kuwada and Yin 1983; Reale and Brugge 1990; McAlpine et al. 2001). When binaural electrical stimulation was directly compared with acoustical stimulation, electrically evoked ITD functions reached their maximum at shorter ITDs (Smith and Delgutte 2007). Some ITD functions determined here demonstrated very sharp "local peaks" near 0 μs (Fig. 5 or 7, asterisks) that were not reported in acoustic stimulation. These could result from the absence of cochlear spontaneous activity and the hypersynchronization of the responses to the electrical stimulus (review in Hartmann and Kral 2004).

**Response Classes and Parameters**

Units with several peaks ("nonclassified") were found in both hearing and deaf animals. Nonclassified (irregular) ITD functions were also described previously in single units (Smith and Delgutte 2007). They might result from integration of activity from different ITD-sensitive cells in subcortical nuclei. It is also possible that they receive inputs from units with "irregular" ITD curves described in the olivary complex (Yin and Chan 1990; Spitzer and Semple 1995; Batra et al. 1997). Nonetheless, the majority of responses showed a "regular" behavior with changing ITD.

Although ITD sensitivity in itself did not require auditory experience for development, the reduced number of responsive units, reduced evoked firing rate, MD and "smoothness" (signal-to-noise ratio) of the ITD functions demonstrate fundamental deficits in cortical ITD sensitivity in CDCs.

**Intensity Dependences**

Auditory deprivation caused most prominent changes in the intensity dependence of the responses. Different units from controls had similar thresholds in relation to EBR thresholds, leading to a population rate-intensity function that very closely followed a sigmoid (Fig. 3e). In CDCs, individual units had different thresholds, leading to a population rate–level function that increased nearly linearly with increasing stimulation intensities. Unit responses in CDCs had a compressed dynamic range. In combination with a decrease in cortical threshold with electric stimulation in CDCs (Kral et al. 2005), these data show a hypersensitivity of responsive cortical units in deafness (ibid.; cf. Kotak et al. 2005). Nonetheless, nonresponsive units were more frequent in CDCs.

With acoustic stimulation, an interaction between stimulus intensity and response latency was observed (Heil 2004), which is known to affect ITD functions. At higher intensities, the response latency decreases and consequently the ITD functions should shift (cf. Irvine et al. 1995). Although some units with
acoustic stimulation have shown this effect (Fig. 5 in Caird and Klinke 1983), the majority of central neurons have not. In general, ITD functions in the inferior colliculus broaden with increasing mean binaural intensities (Yin et al. 1985) but do not show a systematic change in best ITD. In contrast to these studies, we did not observe a general broadening of the ITD function with intensity. Many cortical units displayed a remarkable stability of ITD functions with increasing intensities. However, if there were shifts in ITD_{best} or ITD_{center}, they were nearly always toward smaller ITDs (decreasing with increasing stimulation levels). This effect could behaviorally lead to increasing underestimation of the azimuth with increasing stimulus intensities. The perceptual consequence of increasing mean binaural level on ITD is not known. Based on time-intensity-trading investigations, it is probable that intensity receives more weight with increasing binaural intensity levels (e.g., Simon et al. 1994; Dietz et al. 2009). In the present study, stimuli were delivered at an interaural intensity difference of 0 dB. Increasing binaural intensity in such a condition should favor interaural intensity difference (binaural intensity difference of 0 dB) over ITD, causing a shift of the ITD curve toward midline. Time-intensity-trading in deaf and hearing animals is currently under study in our laboratory.

Release from early inhibition (Carney and Yin 1989) could be the mechanism of the decrease in ITD_{best} and ITD_{center} with increasing binaural intensities. Early inhibition is effective at worst ITD, mainly when the ipsilateral stimulus is leading. For this early inhibition, a precise timing of both inputs is required. To test the possible involvement of inhibition in the results, ITD_{contrast} has been evaluated. Inhibition is responsible for ITD_{contrast} values larger than unity (Nishino et al. 2008), which were found only at low intensities and only in HCs. ITD_{contrast} successively diminished with increasing intensities in HCs, as did MD. This is different from acoustical stimulation (ibid.) and is consistent with the hypothesis that an escape from inhibition occurs with increasing electrical intensities in controls. It is most likely related to hypersynchronization of auditory nerve responses to the electrical stimulus, which increases with increasing intensities (Hartmann and Kral 2004). It would also explain the shift of ITD functions with increasing intensity, as inhibition appears to shift these functions away from 0 µs (Grothe 2003; Pecka et al. 2008).

In deaf animals, ITD functions did not change with increasing stimulation levels. Either the inhibition is not effective even at low intensities in CDCs or time–intensity trading does not operate in CDCs. Due to the compressed dynamic range of...
unit responses in deaf, units go into saturation already close to EBR thresholds. Consequently, in CDCs at threshold intensities phenomena appear that are observed at high intensities in HCs. Thus, an extensive deficit in central processing of stimulus intensity further aggravates the hypersynchronization of the auditory nerve to CI stimulation, leading to perceptual deficits in spatial hearing (van Hoesel and Tyler 2003; Senn et al. 2005).

**Bimodal Responses**

A significant proportion of units had bimodal PSTHs (for acoustic stimulation, see, e.g., Brugge et al. 1969). With acoustic stimulation, both the latencies of first and the second responses are notably longer in the dorsal zone than in A1 (Stecker et al. 2005). Considering the anatomical locations of the recording positions (Fig. 1), units from DZ are not likely to contribute to the present results. Mrsic-Flogel et al. (2005) demonstrated that first and second responses found in A1 have sometimes complementary spatial tuning. This agrees well with the present results. The mechanism behind the second response remains unclear. One possible participating process is a rebound from early inhibition (see also Pecka et al. 2007).

Bimodal cells with complementary tuning can be expected to respond best to subsequent stimuli with “opposite” ITDs (opposite locations in space). In this latency range, the precedence effect (succeeding sounds are mislocalized to the position of their predecessors) has sometimes been related to inhibition in inferior colliculus (Litovsky and Yin 1998a, 1998b; Pollak et al. 2003). Correspondingly, we assume that the cortical complementary responses observed in the present study take part in the processing of reverberating sounds. If a unit responds to both the original sound and the reverberation, both inputs lead to activation of similar cell populations and the stimulus and its reverberation are experienced as a single auditory event. As not all units respond in a bimodal complementary fashion, the finding indicates the existence of specialized network-processing reverberations.

Bimodal responses were rare in CDCs. This indicates that deaf animals, if regaining hearing late in life, might have difficulties in suppressing acoustic reverberations. Indeed, no evidence for the precedence effect has also been demonstrated in CI subjects with prelingual onset of deafness (Agrawal et al. 2006; cf. also Seeber 2007). The present findings represent a possible neuronal correlate to these psychophysical observations.

**Recording Position: Rostral versus Caudal A1**

With electrical stimulation, the excited cochlear partition is not the consequence of the frequency of the stimulus (as with acoustic stimulation) but rather the result of the position of the electrode array within the cochlea. As a rule, neither in human nor in animal cochleae are conventional implants located in the low-frequency region. In the present study, the array was positioned in the cochlear region with characteristic frequencies ~10–30 kHz (e.g., Kral et al. 1998). Thus, with regard to binaural interactions, CIs appear to stimulate the inappropriate cochlear region.

With acoustic stimulation, on the other hand, ITD functions are typically collected for neurons with low characteristic frequencies, typically less than 5 kHz (McAlpine et al. 2001). At higher frequencies, the cochlea is not able to code the fine structure of the afferent signal, and therefore the ITD sensitivity is lost. Nonetheless, the ITD can still be processed, and for frequencies greater than 1500 Hz, sensitivity to ITD in the signal envelope has been observed (Middlebrooks and Green 1990; Griffin et al. 2005). The medial superior olivary complex also contains high-frequency neurons (Guinan et al. 1972; review in Schwartz 1992). The present data demonstrate that the brain indeed can make use of ITD in temporal fine structure even at high-frequency regions. However, the fitting of ITD functions to the templates was significantly better in caudal parts of A1 in controls, indicating a superior extraction of ITD information in cortical low-frequency representation.

With increasing best frequencies, the best delay decreases in acoustic stimulation (McAlpine et al. 2001). We compared ITD_{best} in the rostral and caudal hot spots, keeping in mind that the most rostral recording positions are far off from the region where such relations were demonstrated with acoustic stimulation. In controls, the rostral spot showed smaller ITD_{best} with nonsignificantly different ITD_{center}, indicating steeper ITD functions. In this sense, sensitivity to ITD was more contralateral and the rising slope of ITD functions covered a larger range of ITDs in the caudal spot. This agrees well with the fact that the low-frequency area of field A1 is “experienced” to process and represent ITDs. The difference between the spots was nominally small in HCs, indicating that long-term hearing via binaural CIs in postlingually deaf might further improve the ITD representation in the most rostral part of A1, provided that binaural implants present interaural timing cues consistently. In deaf cats, the difference between the rostral and caudal cortical positions was not apparent, suggesting a deterioration in ITD representation within field A1. Smeared cortical gradients have been reported in the cortical aural representation in CDCs (Kral et al. 2009) and in cochleotopic organization in neonatally deafened cats (Fallon et al. 2009).

**Clinical Consequences**

It remains unclear whether field A1 directly contributes to spatial localization or whether it is a relay station required by other auditory fields for sound source localization. Whichever alternative is correct, the present deficits demonstrate that congenital auditory deprivation has an extensive effect on binaural representation in auditory cortex. As the cortex is essential for auditory spatial localization, this implicates that congenitally deaf subjects would have extensive difficulties in localization of sound sources, especially in reverberating environments and at high intensities.

Indeed, deaf subjects implanted with binaural CIs have difficulties in differentiating ITDs (van Hoesel and Tyler 2003; Senn et al. 2005; Seeber and Fastl 2008). The present findings suggest that central auditory deficits contribute to these difficulties. However, the present data have shown that the elementary extraction of ITD information is rudimentary preserved in the absence of hearing experience. Bilateral implantation during postnatal development and appropriate training might thus help to overcome the deficits, provided the speech processors deliver ITD information consistently and binaural implantation is performed early, within the sensitive period for cortical plasticity (Kral et al. 2002, 2006). Future studies will show whether this is feasible.

**Supplementary Material**

Supplementary material can be found at: http://www.cercor.oxfordjournals.org/
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None declared.

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