

Vision-independent Adjustment of Unit Tuning to Sound Localization Cues in Response to Monaural Occlusion in Developing Owl Optic Tectum

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Neurons in the developing optic tectum adjust their tuning to auditory localization cues in response to chronic monaural occlusion so that auditory spatial fields align with visual receptive fields (VRFs). We tested whether this adaptive adjustment of auditory tuning requires visual instruction. Both eyelids were sutured closed at the same time that one ear was occluded in two barn owls that were 1 month old. After 70 and 100 d, respectively, the tuning of units to interaural level difference (ILD) and to interaural time difference (ITD) was measured. These data were compared with equivalent data from 15 normal owls.

Unit tuning to ITD was shifted from normal in both of the monaurally occluded owls. In one owl, ILD tuning was also clearly shifted. In the other owl, the map of ILD was flipped upside down and adaptive adjustments in ILD tuning could not be assessed. Instead, adjustments in ILD tuning were observed following removal of the earplug with the eyelids kept closed. Unit tuning was monitored at several sites in the tectum for 1 month after earplug removal using chronically implanted electrodes. Then, ILD tuning was resampled across the entire tectum. Both measures indicated shifts in ILD tuning in response to removal of the earplug in the second blind owl. In both animals, the magnitude of the shifts in ILD tuning and ITD tuning was smaller than has been observed previously in monaurally occluded but sighted owls. The results demonstrate that the brain can make adaptive adjustments in ILD and ITD tuning in response to early monaural occlusion even without the guiding influence of vision.

In the optic tectum (superior colliculus), neurons are tuned to the location of visual and/or auditory stimuli, and they are organized according to their spatial tuning to form a multimodal map of space (Knudsen, 1982; King and Palmer, 1983; Middlebrooks and Knudsen, 1984; Wong, 1984; King and Hutchings, 1987). The auditory spatial tuning of these neurons results from their tuning to sound localization cues such as interaural differences in sound level (ILD) and timing (ITD) (Wise and Irvine, 1985; Olsen et al., 1989). Previous studies in barn owls

and ferrets have shown that, during development, auditory spatial tuning adjusts adaptively in response to chronic occlusion of one ear: over a period of weeks or months, neurons become tuned to sound sources at the locations of their VRFs, despite the abnormal auditory cues caused by the earplug (Knudsen, 1985; King et al., 1988). The preceding article demonstrates that the underlying basis of this adaptive adjustment is a systematic compensatory change in the tuning of tectal neurons to values of ILD and ITD (Mogdans and Knudsen, 1992).

A powerful, vision-based mechanism is known to be capable of adjusting the auditory spatial tuning of tectal neurons during development (King et al., 1988; Knudsen and Brainard, 1991). In owls raised with the visual field displaced by prisms, tectal neurons become tuned to sound source locations corresponding to their optically displaced, rather than their normal, visual receptive field (VRF) locations. Obviously, the same vision-based mechanism could underlie the adaptive adjustment of auditory spatial tuning in response to monaural occlusion. However, other potential sources of calibrating information exist, including information contained within auditory signals themselves and information provided by somatosensory and kinesthetic inputs.

Evidence that the brain can use nonvisual information to calibrate certain aspects of auditory spatial tuning comes from a study on owls raised from birth with the eyelids sutured closed (Knudsen et al., 1991). Auditory spatial tuning was found to be abnormal in nearly all portions of the tectum; indeed, in some of the blind birds, the auditory map was flipped upside down. However, in every case, the representation of the region of space directly in front of the animal was normal, indicating that this portion of the map is less dependent on visual calibration than the rest. Because sound originating from this portion of space always gives rise to cue values near the midpoint of the experienced range, it was hypothesized that the brain uses such intrinsic auditory information to help position the neural representation of frontal space correctly in the tectum.

To resolve the issue of whether nonvisual sources of information can contribute to adaptive adjustment of auditory spatial tuning, we studied the effects of simultaneous eyelid closure and monaural occlusion on auditory tectal unit tuning in developing owls. Unlike the study mentioned above (Knudsen et al., 1991), these owls experienced normal vision (and hearing) until 1 month of age when the experiment began. The direction of adjustment we refer to as adaptive is the direction that tends to realign a neuron's auditory receptive field with its VRF. Because an earplug attenuates and delays the auditory signal at the occluded ear (Knudsen et al., 1984), this corresponds, for a left ear oc-

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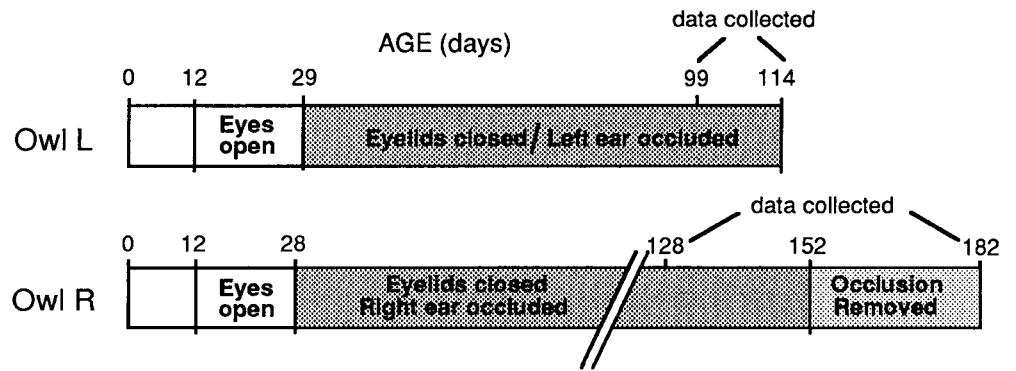


Figure 1. History of sensory experience for experimental owls L (above) and R (below). The age of each bird is indicated in days relative to hatching.

clusion, to a shift toward right-ear-greater ILDs and right-ear-leading ITDs and, for a right ear occlusion, to shifts in the opposite direction. The results demonstrate that indeed, without the aid of vision, the brain can make adaptive adjustments in unit tuning to sound localization cues that, although less extensive than those observed typically in sighted owls, are nonetheless striking.

Materials and Methods

The study is based on data from 17 barn owls (*Tyto alba*): 15 normal and 2 experimental. The data from the normal owls have appeared previously in a normal study and as control data in a study of the effects of monaural occlusion in sighted owls (Olsen et al., 1989; Mogdans and Knudsen, 1992). Data collection and analysis techniques were identical for the normal and experimental owls.

Sensory deprivation and rearing conditions. Baby owls were raised by their parents in an aviary until 18–25 d of age. The owls were removed and placed in rearing boxes in groups of two or three. Three experimental owls were chosen randomly for combined eyelid closure and monaural occlusion; siblings of these owls were raised normally or were monaurally occluded.

When the experimental owls were 28 or 29 d old, they were anesthetized with halothane and nitrous oxide, a foam rubber earplug (E.A.R. Cabot Corp.) was sutured into the external meatus on one side, and the margins of the eyelids were incised and sutured together (Fig. 1). Within 2 weeks, the upper and lower lids of two of the owls had fused to form continuous dermal layers over the eyes. The eyelids of the third owl did not fuse completely. Therefore, physiological data from this owl are not included in this study.

The three experimental owls were raised together but apart from the other owls, because eyelid closure put them at a competitive disadvantage when searching for food (dead mice). These owls were given special attention to ensure that they ate well and grew normally; both exhibited above-average growth curves. In addition, these owls were handled frequently and were exposed to a rich auditory environment. Nevertheless, they were more sedentary than their sighted siblings. At 60 d of age, when the sighted birds began to fly and were placed in a flight aviary, the visually deprived birds would not fly and remained in the rearing box where they continued to receive daily attention.

The two owls with fused eyelids were unable to see anything but large changes in illumination. Owls are normally vigorously hostile to the approach of foreign objects. The two experimental owls did not respond at all to objects brought to within millimeters of the eyelids. When touched by objects, however, they exhibited strong startle responses and threat displays.

The acoustic effects of the earplugs were not measured directly. However, the effects of the identical earplugging procedure on adult owls have been reported previously (Knudsen et al., 1984; Mogdans and Knudsen, 1992). Based on visual inspection, the earplugs in both birds remained firmly in place throughout the experiment.

Neurophysiology. Single units and clusters of two or three units were recorded extracellularly with insulated tungsten microelectrodes from all layers of the optic tectum. Except for the chronic recordings (see below), the techniques and equipment used were the same as those described in detail in a previous study (Olsen et al., 1989). Owls were

anesthetized with intramuscular injections of ketamine HCl (15 mg/kg) and diazepam (5 mg/kg). Experimental owls had their earplugs removed and their eyelids opened. A headpiece was cemented to the skull, and a small craniotomy was made in the skull over the region of the optic tectum. The body was suspended in prone position at the center of an acoustic isolation chamber (IAC 404A) and the head was aligned in the chamber using retinal landmarks (the eyes are essentially stationary in the head). The eyes of the visually deprived owls were found to be oriented normally (unlike those of blind-reared owls: Knudsen, 1989), probably because of the 2 weeks of vision they experienced before eyelid closure (Fig. 1).

Auditory tuning to ILD and ITD was measured by presenting noise-burst stimuli through earphones. The dichotic stimuli consisted of computer-generated noise, digitally high-passed at 4 kHz (so that sound transmission through the interaural canal did not affect the signals over the range of ILDs tested; Moiseff and Konishi, 1981), and transduced by subminiature earphones (Knowles ED-1914) coupled to damping assemblies (Knowles BF-1743). The amplitude of the signal for each channel was controlled with a programmable attenuator. Time delays between the channels were produced by computer-calculated shifts in the waveform of the signal.

For each measurement, 10 series of stimuli were presented. Within each series, sound bursts were 50 msec in duration, delivered at a rate of 1/sec, at an average binaural level of 20 dB above threshold, and the order of ILD or ITD values was randomized. Unit response was quantified as the number of spikes occurring in the 100 msec following stimulus onset minus the number of spikes occurring in the 100 msec prior to the stimulus (spontaneous rate). Best ILD and best ITD were defined as the center of the range of values for which a stimulus produced > 50% of the maximum response. When ILD tuning was measured, ITD was held constant at its best value; when ITD tuning was measured, ILD was held constant at its best value.

VRFs were defined as the area from which stimuli produced an increase in discharge rate. Visual stimuli consisted of bars and spots projected from an ophthalmoscope, imaged on a translucent hemisphere (radius, 57 cm) that was brought into the chamber for visual tests and then was removed. The hemisphere was calibrated in double-pole coordinates (Knudsen, 1982) such that azimuth indicates degrees to the right (R) or left (L) of the owl's midsagittal plane and elevation indicates degrees above (+) or below (–) the owl's visual plane.

At the conclusion of an experiment, antibiotic was applied to the brain surface, the craniotomy was sealed with dental acrylic, the eyelids were sutured closed, the ear was occluded with a fresh plug, and incisions were infused with Xylocaine. The owl recovered from anesthesia in 1–2 hr and was then returned to its home cage.

Restricted unit sampling. Unit sampling was restricted largely to bimodal (auditory-visual) units from the rostral third of the optic tectum, representing visual locations from L20° to R20° in azimuth and from –30° to +20° in elevation, where unit properties are fairly uniform in normal animals (Knudsen, 1984; Olsen et al., 1989). In the frontal region of space represented in this portion of the tectum, ILDs of frequencies greater than 5 kHz vary almost exclusively and monotonically as a function of source elevation (due to a physical asymmetry in the external ears), whereas ITDs vary almost exclusively as a function of source azimuth. Correspondingly, unit best ILDs and best ITDs correlate strongly with their VRF elevations and azimuths, respectively, in normal owls (Olsen et al., 1989). Changes in these relationships were used to detect

abnormal ILD and ITD tuning in the experimental owls. All of the statistical analyses were based only on data obtained from recording sites with VRFs located within this frontal region. In addition, because the preceding companion study revealed that auditory adjustments in sighted birds tend to be larger and more consistent in the tectum ipsilateral to the occluded ear (Mogdans and Knudsen, 1992), recording sites were sampled primarily from the ipsilateral tectum.

Chronic recordings. In order to follow changes in ILD and ITD tuning at specific sites over time, insulated tungsten microelectrodes were cemented in place with dental acrylic in owl R. At 1 week intervals, the owl was anesthetized with halothane and nitrous oxide, the eyelids were opened, the owl was positioned in the acoustic chamber, and auditory and visual tuning at each site was measured as described above. Frequency tuning at each site was also measured based on the number of spikes elicited by tone bursts with 5 msec rise and fall times, presented at 20 dB above broadband threshold, and with best ILD and ITD values. When finished, the eyelids were sutured closed and the owl was removed from the chamber and returned to its home cage.

Results

General unit properties

In the two birds raised with one ear occluded and both eyelids closed, the most conspicuous abnormal response property in the optic tectum was the shift in unit tuning to ILD and ITD, as described below. However, other unit properties were also abnormal: spontaneous activity was low, auditory responses were often weak and erratic, many units could not be driven at all by dichotic stimuli, and others habituated strongly to repetitive stimulation. Nevertheless, among the responsive units, latencies and thresholds were the same as those measured in the normal owls. In contrast to the visual responses reported in blind-reared owls (Knudsen et al., 1991), visual responses in these owls were strong and apparently normal, probably as a result of the initial 2 weeks of normal vision (Fig. 1).

ILD tuning

The sharpness of unit tuning to ILD was largely unaffected by monaural occlusion and eyelid closure (Table 1). The exception was that 6 out of 41 recording sites (15%) in owl R (right ear occlusion) continued to respond to sound levels much greater in the left ear than in the right, resulting in ILD tuning curves that were "open" to left-ear-greater ILDs (referred to as excitatory-inhibitory, or EI, responses in other literature). Among comparable units from normal owls, only 1 unit out of 91 (1%) continued to respond to large left-ear-greater ILDs.

In contrast, the value of ILD to which units were tuned was altered conspicuously, as revealed by comparing the relationship of best ILD (see Materials and Methods) with VRF elevation in normal and experimental animals. The relationship of best ILD with VRF elevation observed in normal owls is shown in Figure 2A. For recording sites in the rostral tectum (see Materials and Methods), best ILD changed continuously and approximately linearly ($r^2 = 0.89$) from 18 dB right-ear-greater for recording sites with VRFs near $+20^\circ$ to left-ear-greater by 15 dB for recording sites with VRFs near -25° . The regression line for the entire data set passed through 0 dB when VRFs were at -5° . Thus, recording sites with VRF elevations above -5° normally were tuned to right-ear-greater ILDs and recording sites with VRF elevations below -5° normally were tuned to left-ear-greater ILDs. (Note that the regression line plotted in Fig. 2A is for the data set restricted to VRFs located between -5° and $+20^\circ$, described below.)

The pattern of best ILDs recorded in owl L (left ear occluded) exhibited two abnormal aspects. First, for recording sites with VRFs located above -5° ("a" in Fig. 2B), which normally are

Table 1. Sharpness of tuning to ILD and ITD^a

	Normal ^b	Owl L	Owl R	Owl R ^c
ILD (50% width, dB)				
mean	12.7	11.1	13.0	10.7
SD	4.3	3.3	4.3	2.9
n	88 ^d	37	35 ^e	28
ITD (50% width, μ sec)				
mean	43.2	38.5	52.0**	35.5
SD	14.9	10.1	17.2	11.8
n	91	37	41	31

^a All recording sites with visual receptive fields centered between $L20^\circ$ and $R20^\circ$ and between -30° and $+20^\circ$.

^b Data from 15 owls.

^c Data collected 30 d after the earplug was removed, during which time the eyelids remained closed.

^d Data from three recording sites that were not included in the mean had ILD tuning curves that were open to one side; two to right-ear-greater, one to left-ear-greater.

^e Data from six recording sites that were not included in the mean had ILD tuning curves that were open on one side; all to left-ear-greater.

** Significantly different from normal (two-tailed *t*-test, $p < 0.01$).

tuned to right-ear-greater ILDs, best ILD varied linearly with VRF elevation ($r^2 = 0.85$), but the values were systematically shifted toward abnormally large right-ear-greater values (the direction of an adaptive adjustment; ANOVA, $p < 0.01$). An estimate of the magnitude of this shift was made from the average difference between the regression lines calculated for data from the normal owls and from owl L, both data sets restricted for VRF elevations between -5° and $+20^\circ$ (Fig. 2A,B). The difference in the regression lines at $+7.5^\circ$ elevation (the midpoint) was 5.3 dB (vertical broken line, Fig. 2B).

The second abnormal aspect to the pattern of best ILDs in owl L was that no recording sites were tuned to large, left-ear-greater values ("b" in Fig. 2B): best ILDs remained at about 0 dB, or even at right-ear-greater values (mean, 2.0 ± 2.5 dB right-ear-greater; $n = 25$) as VRF elevations descended from -5° toward -30° . A consequence of this lack of progression in best ILD values was that the difference in best ILD from the predicted normal value (Fig. 2A) increased with decreasing VRF elevation to as much as 20 dB for recording sites with VRFs near -30° .

Recording sites with VRFs located between -5° and -10° exhibited the smallest shifts in best ILD. The average best ILD in this portion of the tectum was 2.4 ± 2.2 dB right-ear-greater ($n = 11$) in owl L compared with 1.4 ± 3.5 dB left-ear-greater ($n = 21$) in normal owls, indicating a shift of only 4 dB.

An average overall shift in ILD tuning in owl L was calculated from the differences between measured best ILDs and those predicted from the normal regression line. Calculated in this manner, the mean shift was $7.5 \text{ dB} \pm 4.2 \text{ dB}$ ($n = 37$) toward right-ear-greater (one-tailed *t*-test, $p < 0.01$). Note, however, that this calculation ignores the change in pattern of best ILDs across the map and therefore is affected by sampling density, which was not uniform.

The pattern of best ILDs recorded in owl R (right ear occluded) differed fundamentally from the normal pattern. The progression was upside-down: best ILDs changed from large left-ear-greater values for recording sites with VRFs located high, toward right-ear-greater values as VRF locations descended (Fig. 2C). This upside-down orientation of the ILD map,

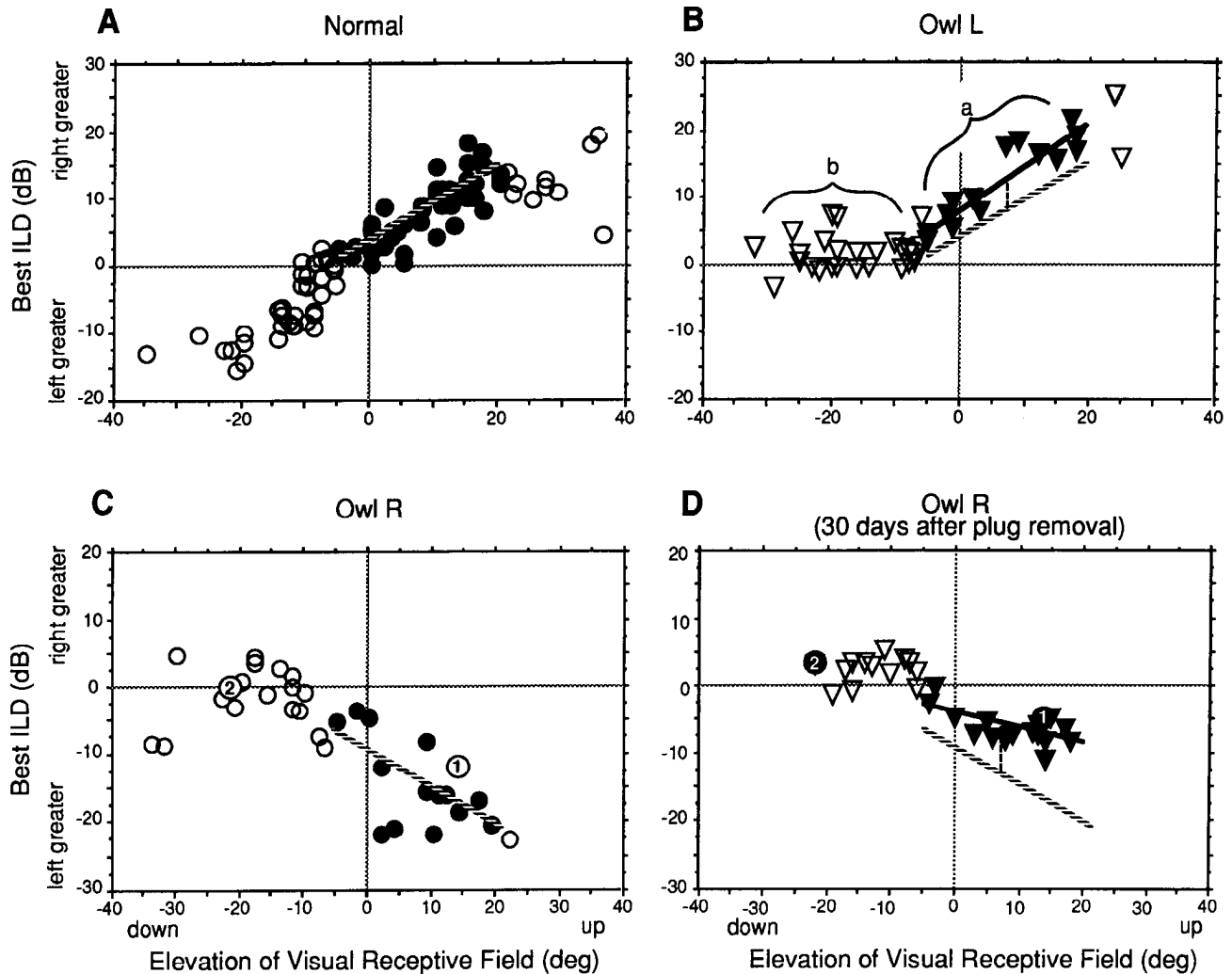


Figure 2. The relationship between best ILD and VRF elevation in the tectum of normal owls and experimental owls. In all cases, the data are restricted to recording sites with VRFs located between L20° and R20°, and with ILD tuning curves that were closed (Table 1). The data for owls L and R (B–D) are from the tectum ipsilateral to the occlusion. *Solid symbols* in each graph indicate data from recording sites restricted to VRF elevations located between -5° and $+20^\circ$ for which, in normal owls, best ILDs vary as a linear function of VRF elevation and have values that are right-ear-greater; *open symbols* indicate data that are outside of this range. **A**, Data from 15 normal owls. The regression calculated for the restricted data set (*broken line*; $y = 0.52x + 3.92$; $r^2 = 0.66$) was used to assess best ILDs measured in the equivalent portion of the tectum of owl L. **B**, Data from the left tectum of owl L, which had the left ear occluded and both eyelids closed. The *solid symbols* indicated by *a* represent the restricted data set. Within this portion of the tectum, ILD tuning changed continuously with VRF elevation. The regression of these data (*solid line*; $y = 0.65x + 8.26$; $r^2 = 0.85$) was shifted toward right-ear-greater by 5.3 dB at its midpoint (*vertical broken line*) relative to the normal regression (*thick broken line*). The *open symbols* indicated by *b* represent data from the ventral portion of the tectum where best ILDs no longer changed with VRF elevation. In normal owls, units in this part of the tectum are tuned to left-ear-greater ILDs. **C**, Data from the right tectum in owl R, which had the right ear occluded and both eyelids closed, collected immediately after earplug removal. The progression of best ILD with VRF elevation is reversed relative to that observed in normal owls. The regression for the data from the restricted set was $y = -0.51x - 9.98$, $r^2 = 0.33$ (*thick broken line*). The *encircled 1* and *2* indicate data from chronically recorded sites 1 and 2, respectively (see Fig. 4). **D**, Data from the right tectum of owl R collected 30 d after the earplug had been removed; the eyelids had been kept closed. The regression for the data from the restricted set (*solid line*; $y = -0.22x - 4.41$; $r^2 = 0.38$) was shifted toward right-ear-greater by 7.5 dB at its midpoint (*vertical broken line*) relative to the regression for comparable data measured immediately after earplug removal (*thick broken line*). *Encircled 1* and *2* indicate data from chronically reported sites 1 and 2, respectively, collected 28 d after earplug removal.

which occurred in both tecta (Fig. 3), is observed frequently in owls raised blind (Knudsen et al., 1991) and is therefore likely to be a result of visual deprivation per se.

Although fascinating in its own right, the upside-down ILD map obscured adaptive adjustment in ILD tuning that might have taken place. If we postulated that, without an earplug, best ILDs would have flipped symmetrically about 0 dB ILD, then the difference between the inverse of the normal regression (Fig. 2A) and the regression of the data from owl R (Fig. 2C) indicated an average adaptive shift of 6.2 dB. However, there is only indirect evidence that this postulate is true (Knudsen et al.,

1991). Therefore, in order to obtain direct evidence of vision-independent adjustments in owl R, we removed the ear occlusion permanently (at 152 d of age; Fig. 1) while keeping the eyelids shut, and measured the changes in ILD tuning that occurred following occlusion removal. These changes were assessed in two ways. First, chronic electrodes were used to measure changes in ILD (and ITD) tuning over a 28 d period at selected tectal sites. Second, the pattern of best ILDs across the tectum was remapped 30 d after removal of the occlusion, and these data were compared with those collected immediately after earplug removal.

Data from the chronic electrodes are shown in Figure 4. The electrodes were placed at the visual representations of R1°, +14° (site 1) and L7°, -22° (site 2) in the right tectum, and at R10°, -24° (site 3) in the left tectum. For each site, ILD tuning was measured on the day of occlusion removal (day 0, open squares), 7 d later (solid circles), and 28 d later (solid triangles). The location of the VRF at each site remained constant, indicating that the electrodes did not move in the tectum during the 28 d period. However, during this period, ILD tuning shifted toward right-ear-greater values at all three sites (Fig. 4A–C). The shift was clearest at sites 1 and 3, where best ILDs changed from 11.5 to 5.5 dB left-ear-greater and from 10.5 to 19.8 dB right-ear-greater, respectively. Changes in both the left-ear-greater cutoffs and right-ear-greater cutoffs of the tuning curves were responsible for the shifts (Fig. 4A,C). At site 2, adjustment was more modest. Best ILD changed by only 3 dB, due solely to an adjustment in the left-ear-greater cutoff (Fig. 4B).

The data collected by remapping the tectum after 30 d (Fig. 2D) agreed with the data from the chronic recordings (numbered symbols in Figs. 2C,D; 3). For recording sites with VRFs located above -5° (solid triangles, Fig. 2D), best ILDs varied with VRF elevation ($r^2 = 0.38$), but the values were systematically shifted toward right-ear-greater values compared with the regression ($r^2 = 0.33$) calculated for the data collected immediately after earplug removal (ANOVA, $p < 0.01$). The difference in the regression lines at +7.5° elevation (the midpoint, Fig. 2D) indicated an average shift of best ILD of 7.8 dB in the direction of right-ear-greater (the direction of adaptive adjustment). This value compares well with the 6.0 dB shift measured with the chronic electrode (site 1) in this portion of the tectum (Fig. 4A).

The range of best ILD values represented in the right tectum was limited (Fig. 2D), as it was in owl L on the left side (Fig. 2B). In owl R, no units were found that were tuned for values of ILD larger than about 5 dB right-ear-greater. This was not true in the left tectum (Fig. 3), where units with VRFs at elevations between -20° and -30° had best ILDs of up to 13 dB right-ear-greater immediately after occlusion removal. Moreover, based on the data collected from the chronic electrode at site 3 (Figs. 3, 4C), units on the right side could adjust their best ILDs to at least 19.8 dB right-ear-greater following occlusion removal. Thus, whatever caused the limit to the range of best ILDs represented in the right tectum of owl R did not similarly affect the range of best ILDs represented in the left tectum.

ITD tuning

The sharpness of ITD tuning was not altered by monaural occlusion and eyelid closure in owl L (Table 1). However, in owl R, ITD tuning was slightly, but significantly ($p < 0.01$), broader than normal immediately after occlusion removal. By 30 d following occlusion removal, ITD tuning had returned to normal sharpness.

The normal relationship between best ITD and VRF azimuth ($r^2 = 0.82$) is shown in Figure 5A. In owls L and R immediately after occlusion removal, the correlation was weaker ($r^2 = 0.19$ and 0.29, respectively, but note the limited range of azimuths that were sampled), and best ITD values generally were shifted in the adaptive direction: toward right-ear-leading in owl L and toward left-ear-leading in owl R (Fig. 5B,C). The average magnitude of this shift was calculated as the mean of the differences of the experimental data from the normal regression (broken lines in Fig. 5). In owl L, best ITDs were shifted an average of $17.0 \pm 8.2 \mu\text{sec}$ ($n = 37$) toward right-ear-leading; in owl R

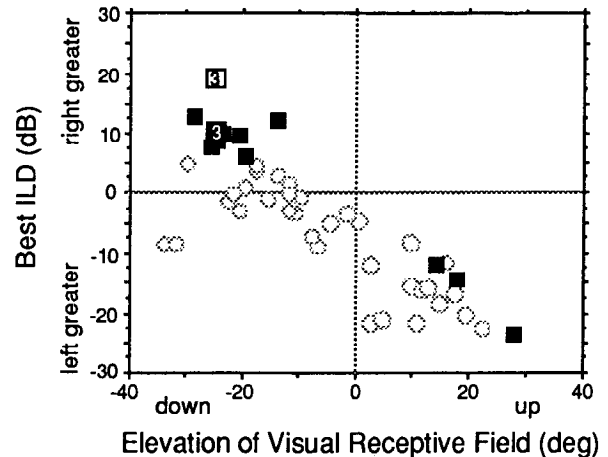


Figure 3. Best ILD plotted as a function of VRF elevation for recording sites in the left and right tecta of owl R measured immediately after earplug removal. *Solid squares* represent data from the left tectum; *open circles* represent data from the right tectum and are same as those plotted in Figure 2C. The symbols marked with a 3 indicate data from the chronically recorded site 3 immediately after (*solid square*) and 28 d after (*open square*) earplug removal (see Fig. 4).

immediately after occlusion removal, best ITDs were shifted $9.7 \pm 20.5 \mu\text{sec}$ ($n = 41$) toward left-ear-leading. A one-tailed t -test indicates that these data were significantly different from normal at the $p < 0.01$ level for owl L and at the $p < 0.05$ level for owl R.

The data collected after 30 d of normal hearing in owl R (Fig. 5D) revealed a strengthened correlation between best ITD and VRF azimuth ($r^2 = 0.77$) and no abnormal bias in the best ITD values (mean, $1.4 \pm 14.2 \mu\text{sec}$; $n = 31$). This implies that a small adaptive shift in ITD tuning had occurred during the 30 d following occlusion removal, although no clear shifts following earplug removal were apparent in the data from the three chronic recording sites (Fig. 4D–F).

Discussion

Adaptive adjustments without vision

The results demonstrate that experience-dependent adjustment of ILD and ITD tuning can occur in the absence of vision. Best ILDs shifted in the adaptive direction both in response to monaural occlusion in owl L and in response to removal of the occlusion in owl R (Figs. 2B,D; 4A–C). The fact that ILD tuning in owl R changed following earplug removal indicates that, in addition to being flipped upside down, the initial correlation of best ILD with VRF elevation (Fig. 2C) did indeed contain an adjustment component as a result of the monaural occlusion. Best ITDs also shifted, on average, in the adaptive direction in response to monaural occlusion and, in owl R, following removal of the occlusion (Fig. 5). This implies that vision-independent mechanisms as well as vision-dependent mechanisms (King et al., 1988; Knudsen and Brainard, 1991) can contribute importantly to the shaping of auditory response properties in the optic tectum during development.

The experiments did not explore the entire range of effects that can result from simultaneous eyelid closure and monaural occlusion. Adjustments to monaural occlusion alone exhibit substantial variability from one individual to the next, even when vision is available to help guide the adjustment process (Mogdans and Knudsen, 1992). The range of variability is in-

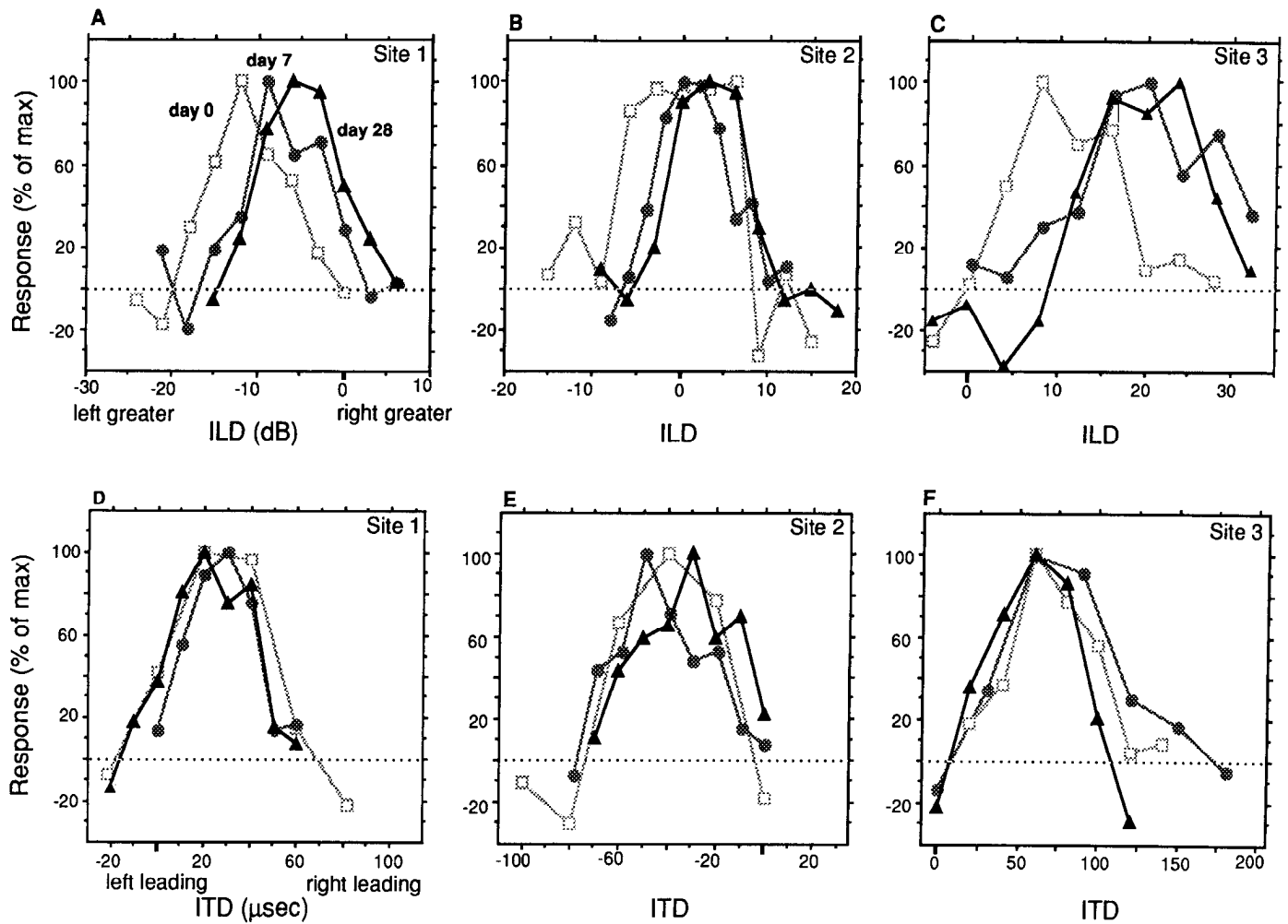


Figure 4. Unit tuning to ILD and to ITD measured in owl R during a 4 week period following earplug removal using electrodes implanted at three selected sites. During this period, the eyelids were kept closed. *A–C* show ILD tuning; *D–F* show ITD tuning. The data represent normalized responses to 30 stimulus presentations of each value, with the order of the values randomized. Negative response values indicate that spike counts were below spontaneous levels. Sites 1 (*A* and *D*) and 2 (*B* and *E*) were located in the right tectum at the visual representation of $R1^\circ$, $+14$ and $L7^\circ$, -22° , respectively. Site 3 (*C* and *F*) was located in the left tectum at the visual representation of $R10^\circ$, -24° . VRF locations did not change during the 28 d period, indicating that the electrodes did not move in the tectum. The data were recorded on the day of earplug removal (*open squares*), 7 d later (*solid circles*), and 28 d later (*solid triangles*). Frequency tuning (best frequency) did not change significantly between day 0 and day 28: site 1, 5.4 and 5.6 kHz; site 2, 5.8 and 6.2 kHz; and site 3, 8.4 and 8.5 kHz, respectively.

creased yet further without vision (Withington-Wray et al., 1990; Knudsen et al., 1991), particularly by the possibility of reversals in the ILD map (Figs. 2*C,D*; 3). Characterization of the range of possible effects would therefore require performing this experiment on a large number of animals. We did not feel the potential results justified such an exhaustive survey. Instead, the data from this study address the more limited question: can the brain make adaptive adjustments in response to monaural occlusion without the aid of vision? The answer is yes.

Magnitude of ILD and ITD adjustments

The magnitude of the adjustments in unit tuning induced in visually deprived owls was smaller than that reported for sighted owls subjected to monaural occlusion (Mogdans and Knudsen, 1992). In that study, the measure used to estimate the size of a shift was the average difference between best ILDs (or ITDs) and the values predicted by the normal regression. Using this measure, the average shift in ILD tuning was 7.5 dB toward right-ear-greater in owl L. In owl R, this measure of best ILD

shift could not be used, because the map of ILD was upside-down. However, the average shift in ILD tuning following earplug removal was 7.8 dB, based on the regression of best ILD on VRF elevation measured immediately after and 30 d after earplug removal (Fig. 2*D*). In sighted owls raised with a monaural occlusion, the average shift in ILD tuning in tecta ipsilateral to the earplug (the side sampled extensively in this study; see Materials and Methods) ranged from 8.7 to 12.5 dB ($n = 6$), with the median being 11.0 dB (Mogdans and Knudsen, 1992). The relative magnitude of adjustment in ITD tuning in the visually deprived owls was even smaller: best ITDs shifted, on average, by only 17 μ sec in owl L and by only 10 μ sec in owl R (Fig. 5), whereas in six monaurally occluded but sighted owls, the average shift in the ipsilateral tectum ranged from 30 μ sec to 63 μ sec, with the median being 47 μ sec (Mogdans and Knudsen, 1992).

The method of monaural occlusion, the rearing conditions, and the care of the animals were identical for the sighted and visually deprived owls in this and the preceding study (Mogdans

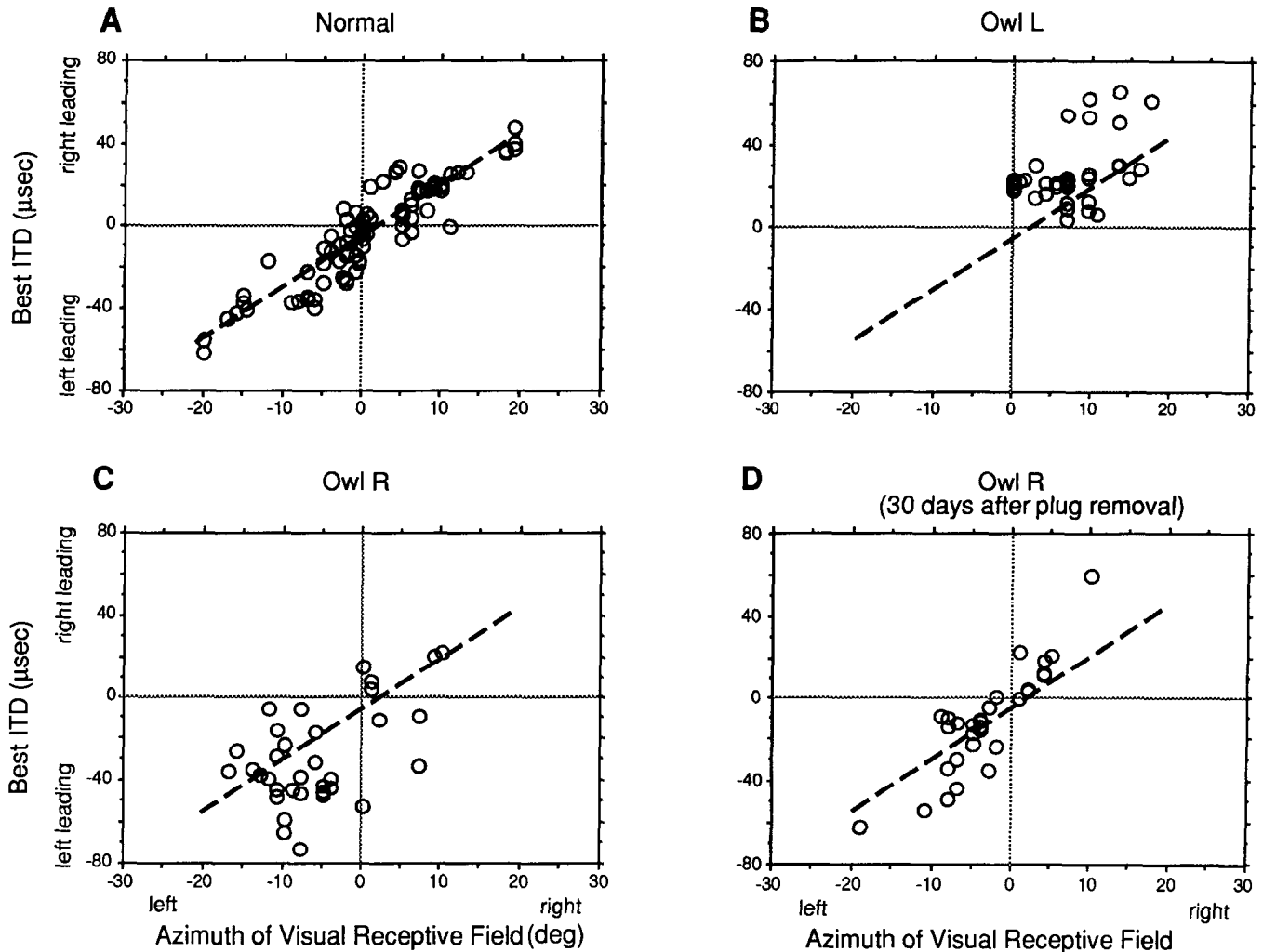


Figure 5. The relationship between best ITD and VRF azimuth in the tecta of normal owls and experimental owls. In all cases, the data are restricted to recording sites with VRFs located between -30° and $+20^\circ$. The data for owls L and R (B–D) are from the tectum ipsilateral to the occlusion. *A*, Data from 15 normal owls. The regression line calculated for these data ($y = 2.5x - 3.95$; $r^2 = 0.82$) was used to assess best ITDs measured in the experimental owls and appears as a *broken line* in each of the plots. *B*, Data from the left tectum of owl L, which had the left ear occluded and both eyelids closed. *C*, Data from the right tectum of owl R, which had its right ear occluded and both eyelids closed, collected immediately after carplug removal. *D*, Data from the right tectum of owl R collected 30 d after the carplug had been removed; the eyelids had been kept closed.

and Knudsen, 1992). In fact, two of the owls in the previous study (owls R3 and L3, with best ILD shifts of 9.8 and 12.5 dB and best ITD shifts of 55 and 57 μsec , respectively) were siblings of owl L in this study. However, the experience of the sighted owls was much richer than that of the visually deprived owls, especially once the sighted owls began to fly at about 60 d of age (see Materials and Methods). Although the visually deprived owls interacted vigorously with each other, sought food, and were handled frequently, they were relatively sedentary and apparently never flew in the aviary. Thus, either vision per se or richer experience seems necessary to adjust unit tuning to its fullest extent.

The difference in the magnitude of the changes in ITD tuning versus ILD tuning was striking: best ITDs changed by an average of only about 20% of the change observed in sighted owls, whereas best ILDs changed by about 75% of the amount observed in sighted owls. This relative difference in the plasticity of ILD versus ITD tuning, which is also apparent in the data from the chronic electrodes (Fig. 4), suggests that by 4 weeks of age (the

age when ears were occluded), ITD tuning is relatively more resistant to change. A possible explanation is that by this age, owls have already experienced azimuth-dependent ITD cues close to the adult range, because the skull is nearly full grown (Knudsen et al., 1984). In contrast, owls have not experienced strong elevation-dependent ILD cues, because these depend on the sound-collecting surfaces of the external ears (facial ruff), which have just begun to grow. Since experience with an adult range of ITD values begins at a much earlier age than does experience with an adult range of ILD values, the process of consolidating unit tuning for ITD might begin earlier, and consequently, changing ITD tuning may require stronger instructional influences beyond this age. This interpretation implies that the sensitive periods when abnormal sensory experience can alter ILD and ITD tuning may be different.

Visual deprivation allows reversal of the ILD progression

Simultaneous eyelid closure and monaural occlusion had an unanticipated effect on ILD tuning: the ILD map in one owl

flipped upside down. Upside-down maps of ILD have been observed previously in blind-reared owls, and the conclusion was drawn that the orientation of the ILD map requires visual instruction (Knudsen et al., 1991). In that study, the eyes were kept closed from birth, whereas in this study the owls experienced 2 weeks of normal vision and hearing before the eyelids were closed. This implies that any vertical disparity in the directional sensitivities of the ears that exists by 28 d of age is not sufficient for vision to establish the correct orientation of the ILD map. Thus, the orientation of the ILD map appears to remain unspecified into the second month after hatching. More important to the issues being considered here is that some of the changes in unit tuning observed in this study may result specifically from visual deprivation rather than from adaptive adjustment.

Plateaus in the ILD map

Over large portions of the tectum ipsilateral to the occluded ear, units that should have been tuned to relatively large ipsi-ear-greater ILDs were limited to ipsi-ear-greater ILDs of less than 3 dB in owl L and less than 5 dB in owl R (open symbols in Fig. 2*B–D*). Abnormal plateauing of the progression of best ILDs occurs also in the tecta of monaurally occluded but sighted owls (Mogdans and Knudsen, 1992), indicating that this effect is caused by the earplug. Although plateaus in the progression of best ILD do not always occur (such as in the left tectum of owl R, Fig. 3), when they do occur, they usually affect the portion of the ILD map that should have represented occluded-ear-greater ILDs. Moreover, the value of ILD represented in the plateau region is always near 0 dB. We have no satisfying explanation for why monaural occlusion should sometimes cause plateauing of the progression of Best ILDs.

Sites of adjustment

It is possible, and even likely, that monaural occlusion affects the processing of localization cues at many levels in the auditory pathway. Had the adjustments been due entirely to changes in the gain and phase response of the monaural signals, then the magnitude of adjustment should have been constant across the ILD and ITD maps. The results demonstrate, to the contrary, that the magnitude and pattern of adjustment, particularly of ILD tuning, varied considerably within and across tecta. For example, within the right tectum of owl R, recording sites with VRF elevations located between $+10^\circ$ and $+20^\circ$ adjusted ILD tuning by an average of 10.1 dB following earplug removal, whereas recording sites with VRFs located below -10° adjusted ILD tuning by an average of only 1.3 dB (Fig. 2*D*). Similarly dramatic differences in the magnitude of ILD adjustments within the tectum of owl L are apparent from the degree to which the relationship of best ILD with VRF elevation (Fig. 2*B*) deviates from a parallel shift of the normal regression (Fig. 2*A*). The magnitude of adjustment in ILD tuning also differed greatly between the right and left tecta in owl R: recording sites with VRFs located below -10° had best ILDs near 0 dB in the right tectum, whereas recording sites with similarly located VRFs had best ILDs near 8 dB right-ear-greater in the left tectum (Fig. 3). Also, during the 30 d following earplug removal, units in this ventral portion of the left tectum (site 3 in Figs. 3, 4) adjusted ILD tuning by 9.3 dB toward right-ear-greater, whereas units in the same portion of the right tectum adjusted ILD tuning by an average of only 1.3 dB toward right-ear-greater (open symbols in Fig. 2*C, D*).

Differential adjustment of ILD tuning across the tectum occurs also in sighted owls, and the implications of differential adjustment for the site of plasticity, discussed in detail for sighted owls (Mogdans and Knudsen, 1992), hold as well for blind owls. In short, differential adjustments imply alterations in ILD coding occurring at or beyond the site in the auditory pathway where interaural level differences are compared, which means at or beyond the level of the posterior division of the ventral nucleus of the lateral lemniscus (VLVp; equivalent to the lateral superior olive in mammals) (Moiseff and Konishi, 1983; Manley et al., 1988).

Potential sources of instructional information

The results demonstrate that the brain is able to adjust unit tuning in response to monaural occlusion without access to visual spatial information. How is this accomplished? One possibility is that spatial information derived from somatic sensation is used to help calibrate the auditory map. Somatic sensation combined with proprioception provides animals with crude information about the location of objects in proximal space (Lackner, 1981; Lackner and Shenker, 1985), and therefore a potential means of associating sound sources (and localization cues) with directions. Another mechanism that relies ultimately on somatosensory feedback is based on audio-motor performance. In response to a sound, the auditory system could make a best guess about the location of the source, and use that guess to guide a strike at the source. Assuming that initial guesses are accurate enough for the animal to succeed in finding the source occasionally, the interpretation of cue values could be adjusted adaptively. However, because owls do not generally explore their environments with their appendages, and because blind owls are particularly passive, it seems unlikely that these mechanisms were responsible for the adaptive changes reported in this study.

Another potential source of auditory spatial information is available entirely within auditory channels. Monaural occlusion shifts the values of binaural cues associated with each location in space, but does not substantially alter the rate or direction of change of cue values with source location. Consistencies in the spatial patterns of cues provide information that could be exploited to compensate for monaural occlusion. For example, although monaural occlusion shifts the range of cue values experienced by an animal, the *center* of that range will always correspond to the region of space directly in front of the animal. A genetically determined connection between neurons encoding the center of the ILD range and the representation of 0° , -10° in the space map was hypothesized in a study of blind-reared owls (Knudsen et al., 1991) to account for the consistently accurate registration of this portion of the auditory space map in the absence of vision. Assuming that such a predetermined connection exists, an adaptive shift would result if structures involved in binaural comparison were simply to shift the range of cue values encoded so that it matched the range experienced by the animal.

All of these potential sources of information may contribute to the establishment of an accurate auditory space map, although our intuition is that experience-based adjustment of the encoded range of cue values is likely to be the most important nonvisual shaping influence. In sighted owls, vision provides additional and considerably more precise information, enabling these and probably other adaptive mechanisms to operate with much greater effect on the auditory map of space.

References

- King AJ, Hutchings ME (1987) Spatial response properties of acoustically responsive neurons in the superior colliculus of the ferret: a map of auditory space. *J Neurophysiol* 57:596–624.
- King AJ, Palmer AR (1983) Cells responsive to free-field auditory stimuli in guinea-pig superior colliculus: distribution and response properties. *J Physiol (Lond)* 342:361–381.
- King AJ, Hutchings ME, Moore DR, Blakemore C (1988) Developmental plasticity in the visual and auditory representations in the mammalian superior colliculus. *Nature* 332:73–76.
- Knudsen EI (1982) Auditory and visual maps of space in the optic tectum of the owl. *J Neurosci* 2:1177–1194.
- Knudsen EI (1984) Auditory properties of space-tuned units in the owl's optic tectum. *J Neurophysiol* 52:709–723.
- Knudsen EI (1985) Experience alters the spatial tuning of auditory units in the optic tectum during a sensitive period in the barn owl. *J Neurosci* 5:3094–3109.
- Knudsen EI (1989) Fused binocular vision is required for development of proper eye alignment in barn owls. *Vis Neurosci* 2:35–40.
- Knudsen EI, Brainard MS (1991) Visual instruction of the neural map of auditory space in the developing optic tectum. *Science* 253:85–87.
- Knudsen EI, Esterly SD, Knudsen PF (1984) Monaural occlusion alters sound localization during a sensitive period in the barn owl. *J Neurosci* 4:1001–1011.
- Knudsen EI, Esterly SD, du Lac S (1991) Stretched and upside-down maps of auditory space in the optic tectum of blind-reared owls; acoustic basis and behavioral correlates. *J Neurosci* 11:1727–1747.
- Lackner JR (1981) Some aspects of sensory-motor control and adaptation in man. In: *Intersensory perception and sensory integration* (Walk RD, Pisk HL, eds). New York: Plenum, pp 143–173.
- Lackner JR, Shenker B (1985) Proprioceptive influences on auditory and visual spatial localization. *J Neurosci* 5:579–583.
- Manley GA, Koepl C, Konishi M (1988) A neural map of interaural intensity difference in the brainstem of the barn owl *Tyto alba*. *J Neurosci* 8:2665–2676.
- Middlebrooks JC, Knudsen EI (1984) A neural code for auditory space in the cat's superior colliculus. *J Neurosci* 4:2621–2634.
- Mogdans J, Knudsen EI (1992) Adaptive adjustment of unit tuning to sound localization cues in response to monaural occlusion in developing owl optic tectum. *J Neurosci* 12:3473–3484.
- Moiseff A, Konishi M (1981) The owl's interaural pathway is not involved in sound localization. *J Comp Physiol* 144:299–304.
- Moiseff A, Konishi M (1983) Binaural characteristics of units in the owl's brainstem auditory pathway: precursors of restricted spatial receptive fields. *J Neurosci* 3:2553–2562.
- Olsen JF, Knudsen EI, Esterly SD (1989) Neural maps of interaural time and intensity differences in the optic tectum of the barn owl. *J Neurosci* 9:2591–2605.
- Wise LZ, Irvine DRF (1985) Topographic organization of interaural intensity difference sensitivity in deep layers of cat superior colliculus: implications for auditory spatial representation. *J Neurophysiol* 54:185–211.
- Withington-Wray DJ, Binns KE, Keating MJ (1990) The maturation of the superior collicular map of auditory space in the guinea pig is disrupted by developmental visual deprivation. *Eur J Neurosci* 2:682–692.
- Wong D (1984) Spatial tuning of auditory neurons in the superior colliculus of the echolocating bat, *Myotis lucifugus*. *Hearing Res* 16:261–270.