Competition and convergence between auditory and cross-modal visual inputs to primary auditory cortical areas

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Submitted 3 May 2010; accepted in final form 25 January 2011

Mao YT, Hua TM, Pallas SL. Competition and convergence between auditory and cross-modal visual inputs to primary auditory cortical areas. J Neurophysiol 105: 1558–1573, 2011. First published January 27, 2011; doi:10.1152/jn.00407.2010.—Sensory neocortex is capable of considerable plasticity after sensory deprivation or damage to input pathways, especially early in development. Although plasticity can often be restorative, sometimes novel, ectopic inputs invade the affected cortical area. Invading inputs from other sensory modalities may compromise the original function or even take over, imposing a new function and preventing recovery. Using ferrets whose retinal axons were rerouted into auditory thalamus at birth, we were able to examine the effect of varying the degree of ectopic, cross-modal input on reorganization of developing auditory cortex. In particular, we assayed whether the invading visual inputs and the existing auditory inputs competed for or shared postsynaptic targets and whether the convergence of input modalities would induce multisensory processing. We demonstrate that although the cross-modal inputs create new visual neurons in auditory cortex, some auditory processing remains. The degree of damage to auditory input to the medial geniculate nucleus was directly related to the proportion of visual neurons in auditory cortex, suggesting that the visual and residual auditory inputs compete for cortical territory. Visual neurons were not segregated from auditory neurons but shared target space even on individual target cells, substantially increasing the proportion of multisensory neurons. Thus spatial convergence of visual and auditory input modalities may be sufficient to expand multisensory representations. Together these findings argue that early, patterned visual activity does not drive segregation of visual and auditory afferents and suggest that auditory function might be compromised by converging visual inputs. These results indicate possible ways in which multisensory cortical areas may form during development and evolution. They also suggest that rehabilitative strategies designed to promote recovery of function after sensory deprivation or damage need to take into account that sensory cortex may become substantially more multisensory after alteration of its input during development.

cross-modal plasticity; sensory substitution; cortical development; traumatic brain injury; stroke

LOSS OF SENSORY DRIVE. Whether as a result of sensory deprivation or brain damage, can result in cortical plasticity, especially early in development. The changes in circuitry that occur as a result of this reactive plasticity may help to restore function or may instead prevent restoration of normal function. Although cortical plasticity can involve either intramodal or cross-modal plasticity, interference with normal function seems more likely to occur as a result of ectopic, cross-modal invasion of the deafenerend structure. For example, loss of visual input can lead to auditory activation of visual cortex (Yaka et al. 2000), and loss of auditory input can lead to cross-modal activation of the understimulated auditory cortex by somatosensory or visual inputs (Allman et al. 2009; Bavelier and Neville 2002; Fine et al. 2005; Finney et al. 2001; Hunt et al. 2006; Lomber et al. 2010; Neville 1990; Neville et al. 1983; Nishimura et al. 2000; Sharma et al. 2007; Sterr et al. 2003). Cross-modal plasticity is known to interfere with the effectiveness of subsequently implanted cochlear prostheses in humans (Lee et al. 2001; see Sharma et al. 2009 for review). To promote restorative plasticity after sensory inputs are compromised and to minimize interference from cross-modal inputs, it would be advantageous to understand how information from existing and ectopic inputs is coordinated and, in particular, how cross-modal inputs affect the amount of territory devoted to the processing of the normal inputs.

Previous studies of sensory deafferentation or deprivation using animal models have examined cross-modal plasticity mainly from the perspective of a complete loss of normal input, such as bilateral enucleation or deafening by cochlear ablation. The disadvantage of using blind or deaf animals to study reactive plasticity in sensory cortex is that the original sensory modality can no longer be activated, preventing examination of how ectopic, cross-modal input affects recovery of the original function, whether through natural means or by implantation of a sensory prosthesis.

In this study we instead employed an approach that brings both normal and cross-modal information to sensory cortex from birth. We tested the hypothesis that cross-modal inputs compete with normal inputs for cortical space. One possible outcome of competition is that the smaller or less active input modality might be suppressed or taken over by the other. Alternatively, segregation of neurons with different response modalities might occur, reducing cortical space available to each. A third possible outcome is that the cross-modal and the normal functions might coexist within the same cortical territory, expanding multisensory processing or perhaps even converging onto single, multisensory neurons.

Using partial deafferentation of auditory thalamus in neonatal ferrets to examine how establishment of auditory cortical territory is affected by invasion of cross-modal, visual information, we found that, in addition to visual and auditory responses, multisensory responses were present at a rate much higher than that seen in normal auditory cortex. These three response types were not spatially segregated, suggesting that information carried by ectopic visual inputs is not sufficient to induce segregation. Our results demonstrate that primary auditory cortex can support both the original auditory and the novel visual function after recovery from damage to afferent
pathways and that multisensory function can be induced simply by experimental convergence of two unisensory inputs. The results provide insight into how multisensory cortex is created on developmental and evolutionary time scales. In addition, our findings provide a more thorough understanding of the reorganization of an affected brain area after recovery from sensory damage or deprivation and have important implications for rehabilitative strategies in patients with damage to sensory pathways.

Preliminary results from some of these experiments have been published previously in abstract form (Mao et al. 2007).

**MATERIALS AND METHODS**

Partial deafferentation of auditory cortex (AC) and invasion of ectopic visual inputs can be produced in ferret kits if retinal axons are induced to invade auditory thalamus (medial geniculate nucleus, MGN) as a result of neonatal midbrain lesions (Sur et al. 1988). It has been shown that the cross-modal auditory cortex (XMAC) in similarly manipulated animals contains functional visual neurons (Roe et al. 1992; Sur et al. 1988; von Melchner et al. 2000). Auditory and multisensory responses were not reported in these previous studies, perhaps because the aim was a complete deafferentation of MGN.

**Animals**

Data were obtained from 25 pigmented ferrets (*Mustela putorius furo*) ages 4 mo or older [ferrets reach full brain and body size at 4 mo (Fox and Bell 1998)]. Timed pregnant ferrets were obtained from Marshall Farms (North Rose, NY) 2 wk before parturition. Nursing dams and kits were fed a high-fat diet and kept on a 14:10-h light-dark cycle. Kits were weaned at 6–8 wk of age. Normal ferrets were either obtained from Marshall Farms as adults or bred in our colony. Nonlactating ferrets were fed Marshall Farms ferret diet and kept on a 12:12-h light-dark cycle. Both male and female ferrets were included in the study. All protocols were approved by the Institutional Animal Care and Use Committee at Georgia State University and met or exceeded standards of care established by the USDA and the Society for Neuroscience.

**Neonatal Surgery**

Surgical procedures were similar to those described previously (Pallas et al. 1999). Ferret kits were manipulated within 24 h after birth. They were anesthetized with isoflurane (1–4%). All surgeries were performed under sterile conditions. After a kit was anesthetized, the skull over the midbrain was exposed and removed with a scalpel. The left superior colliculus and one or both inferior colliculi were then lesioned to varying extents with a heat cautery, and the brachium of the left inferior colliculus was severed with a scalpel blade. The incision was closed using either 6-0 prolene or surgical adhesive (VetBond; 3M, St. Paul, MN). After surgery, the kits were given subcutaneous fluids and a respiratory stimulant (doxapram, 2 mg/kg sq) and warmed under an incandescent lamp. Kits were observed carefully until they recovered from anesthesia and were returned to their dam after they became ambulatory. Analgesics (buprenorphine, 0.05–0.1 mg/kg bid) were given as needed to prevent postoperative pain.

**Preparation for Electrophysiology**

Electrophysiology experiments were done once ferrets reached adult size (>4 mo of age). Before induction of anesthesia, atropine (0.4 mg/kg sq) and doxapram (2 mg/kg sq) were given to counteract bradycardia and to reduce mucosal secretions. Anesthesia was induced by ketamine (40 mg/kg im) and diazepam (2 mg/kg im) or by ketamine (40 mg/kg im) and medetomidine (0.08 mg/kg im). Dexamethasone (1 mg/kg im) was given every 24 h to prevent brain swelling. Animals were intubated, and the cephalic or femoral vein was cannulated. Anesthesia was maintained with an intravenous solution of medetomidine (0.022 mg·kg⁻¹·h⁻¹) and ketamine (5 mg·kg⁻¹·h⁻¹) in lactated Ringer with 5% dextrose (Bizley and King 2008; Bizley et al. 2005). Atropine (0.06 mg·kg⁻¹·h⁻¹ sq) was given as necessary to counteract the bradycardia caused by medetomidine. Animals were artificially respired using a small animal ventilator (SAR 830/P ventilator; CWE, Ardmore, PA). Vital signs including ECG, respiration rate, muscle tone, withdrawal reflexes, end-tidal CO₂, and oxygen saturation as measured by pulse oximetry (SpO₂) were monitored during the surgery and recordings to ensure maintenance of adequate anesthesia. Body temperature was maintained at 38°C with a heating pad. Pups were diluted with atropine ophthalmic drops. Eyes were kept moist with commercial artificial tears solution and protected with custom plano contact lenses (Conforma, Norfolk, VA). The head was stabilized in a stereotaxic device. After the scalp overlying AC was incised and the muscle was retracted from the skull, two burr holes (at coordinates A5.5 ± L1.5) were drilled for optic chiasm recording/stimulation electrodes. Two tungsten rods were cemented to Teflon insulation (diameter 0.008 in. bare, 0.011 in. coated; A-M Systems, Carlsborg, WA) connected to a preamplifier were lowered (~8–10 mm) while responses to strobe light stimulation were recorded until a depth yielding strong visual responses was reached. These tungsten rods were then cemented to the skull and connected to a stimulus isolation unit (BAK Electronics, Mount Airy, MD). An ~0.8 to 1.0-cm-diameter craniotomy was drilled over the AC, and the suprasylvian and pseudosylvian sulci were exposed.

**Recording sites.** The ferret AC is located on the middle ectosylvian gyrus, bounded above by the anterior and posterior arms of the suprasylvian sulcus and below by the pseudosylvian sulcus (Kelly et al. 1986). In this study, recording penetrations were targeted to the primary auditory cortex (A1) and the anterior auditory field (AAF) (Bizley et al. 2005) and were unlikely to impinge on multisensory regions of the sulci surrounding the AC for several reasons. First, we avoided the regions close to the sulci. In addition, our recording depths were generally very superficial given that we recorded the first unit in each electrode pass and then moved on to the next location in the map. Furthermore, the electrode angle combined with the shape of AC as an inverted parabola is such that increasing recording depth would move away from, and not toward, the banks of the sulci. These factors together argue that it is very unlikely that our recording sites were in the suprasylvian territory, although we cannot exclude the possibility. We also cannot exclude the possibility that a few of our recording sites were located in the posterior ectosylvian gyrus, where higher level auditory fields have been described (Bizley et al. 2005). The dura was removed, and AC was covered by either sterile saline or 2% agar (Fisher Scientific, Fair Lawn, NJ) in sterile saline to protect its surface from desiccation. The right side of the skull was cleaned of tissue, and a metal bracket was cemented on the skull to hold it in position. The right ear bar was then released to allow access to the ear for auditory stimulation.

**Extracellular Recording**

The cortical surface was photographed with a digital camera to record the location of each recording electrode penetration. A glass-coated tungsten microelectrode (1–2 MO; FHC, Bowdoin, ME) was used to investigate neuronal activity in AC in response to auditory and visual stimuli. Penetration locations were chosen to sample randomly from as many sites within AC as possible while avoiding sulci and blood vessels. The electrode was lowered in 5-μm steps up to 2,000 μm under the pial surface using a hydraulic microdrive (Kopf Instruments, Tujunga, CA). Once the first unit in each electrode pass was isolated and characterized, another recording location was selected. Most units were isolated within 800 μm of the pial surface.
Sensory stimuli. For each penetration, bars of light from a pantoscope and white noise from a loudspeaker were used to search for responsive neurons. The loudspeaker was placed at a 45° angle between the right side and the front of the animal at a distance of ~10 cm. Auditory searching stimuli were white noise bursts (5-ms ramp, 40–100 ms in duration) with a sound intensity of 60–80 dB sound pressure level, as measured by a sound level meter (model 407764; Extech Instruments, Waltham, MA). After a responsive neuron was found, computer-generated auditory (noise or tones) and visual stimuli (moving or flashing bars of light) were used for testing responses. The speaker was replaced with a calibrated earphone. The earphone was placed in the pinna at the entrance to the ear canal and used to generate noise bursts or pure tone sounds in closed field (ER-2 insert earphone; Etymotic Research, Elk Grove Village, IL). The earphone was calibrated with a microphone (ER-7C probe microphone system; Etymotic Research) using Sigcal software (Tucker-Davis Technologies, Alachua, FL). The normalized file generated by Sigcal was used to correct any nonlinearities in the earphone output when sound was presented. For assessing the responses of single units isolated in each penetration, auditory stimuli were generated by TDT System II hardware and software (Tucker-Davis Technologies), and visual stimuli were synthesized and delivered by a VSG card (Cambridge Research Systems, Rochester, UK). Light stimuli included bars or gratings at eight orientations moving in either direction, presented on a computer screen ~40 cm from the eyes. Bipolar electrical stimulation of the optic chiasm was applied (single pulses at 0.5–1 mA, 60 ms in duration) in addition to light stimuli.

Electrophysiological Data Analysis

Neural responses were amplified (5,000 to 10,000 times; BAK Electronics), filtered (500 Hz to 5 kHz), and monitored on a digital oscilloscope (Hameg Instruments, Mainhausen, Germany). Responses to 10–15 stimulus presentations were gathered from each recording site and digitized at 25 kHz. The evoked responses were averaged and normalized to a sample of spontaneous activity recorded 50 ms before each trial. The recording continued for 1–2 days, after which the animal was deeply anesthetized with pentobarbital sodium (65 mg/kg) for humane euthanasia and harvesting of brain tissue for histological examination.

For each electrophysiological data point, Brainware software (Tucker-Davis Technologies) was used off-line to isolate extracellularly recorded spikes derived from single neurons by their waveform. Artifact rejection was set in Brainware to extract biphasic action potential candidates with both peaks exceeding background noise level. Spikes with similar shape and duration were shown as clusters in the data-sorting window. Poststimulus time histograms (PSTHs) of the selected single units were generated using the same software package. The mean and standard error of the number of spikes to each stimulus presentation were calculated after spontaneous activity was subtracted. Response latencies were determined by the time between stimulus presentation and the time of the first bin in the PSTH that reached at least 20% above background firing rate. Multisensory units were defined as either neurons that responded to both visual and auditory stimuli or neurons that only responded to one modality but could be significantly modulated by stimulation with the other modality (Stein et al. 1993). Statistical significance was determined by comparing the number of spikes per sweep (obtained from the PSTHs) as a response to different stimulus modalities using Student’s t-test (P < 0.05). The proportion of response types was compared across groups.

For calculating the spatial distribution of response types, the area of each AC was normalized to a standard circle with a radius of 1. The locations of recorded units were reconstructed on this normalized AC. To analyze the distribution of different neuron types in AC, we divided AC into four quadrants numbered 1 to 4 (Q1–Q4) as shown in Fig. 1. The quadrants were not intended to correspond to particular auditory cortical areas, although Q2 and Q3 overlap more with the AAF and Q1 and Q4 overlap more with A1. Two lines were drawn along the anterior and posterior arms of the suprasylvian sulcus to form angle A. The third line was drawn just above the tip of the suprasylvian sulcus and perpendicular to the dividing line of angle A. The center of the internally tangent circle (point 0) was defined as the intersection of the dividing lines of angle A and angle B.

Because the shape of the AC in each individual is unique and the location of recording sites differed somewhat across animals, we examined whether pooling data from different animals into one polar plot would bias the data. We performed a heterogeneity χ² analysis to test the homogeneity of data from each group. Heterogeneity χ² is a statistical test based on the premise that if the samples are homogeneous, then the value of χ² should be close to the value of χ² pooled. Therefore, the heterogeneity χ² value is designated χ² heter. The null hypothesis should be rejected if there is a large χ² heter (for details, see Sheskin 2004). If the χ² pooled from each sample is not significantly different from the value of χ² pooled (P > 0.05), χ² heter will be small and the data can be grouped. Applied to our data, the test showed that the electrode penetrations in the four quadrants of normal AC were homogenously distributed (P > 0.05). The same was true for small-lesion and large-lesion groups (P > 0.05). Therefore, data from all ACs in each group were pooled into one polar plot. A χ² analysis was then applied to determine whether recorded neurons were randomly distributed across quadrants independently of their response type. In cases where the distribution was not random (P < 0.05), an analysis of residuals (R value) was calculated to show which quadrants contained the unexpected distribution.

To examine whether neurons with similar responses were clustered, we calculated the average distance between recording sites by translating X and Y values obtained from normalizing AC to polar coordinates on the standard circle using Microsoft Access database software. The distance between each pair of single units was calculated and exported to a spreadsheet. The average distances from each single unit to other auditory, visual, or multisensory units were calculated.
We then compared the mean of average distance between pairs across groups. Electrophysiological data were statistically analyzed using Sigma-stat software (Systat Software, Chicago, IL) and plotted with Sigma-plot (Systat Software). A one-way ANOVA for multiple groups was used. A Tukey’s post hoc test was used for groups that had uneven numbers, and a Fisher least significant difference (LSD) post hoc test was used for groups that had even numbers. A Mann-Whitney U-test for nonnormally distributed data was used for two-group comparisons. Values are means ± SE throughout.

**Assessment of Lesion Size**

*Magnetic resonance imaging scanning.* Magnetic resonance imaging (MRI) was performed in some lesioned ferrets to obtain an assessment of the midbrain lesions before electrophysiological recording. Atropine (0.4 mg/kg sq) and doxapram (2 mg/kg sq) were given 5 min before sedation. Medetomidine (1 mg/kg im) and diazepam (2 mg/kg im) were then given to sedate the animal. Animals were put into an MRI cradle with a heating pad underneath to maintain body temperature. End-tidal CO₂, SpO₂, pulse rate, respiration rate, and body temperature were monitored during the entire process. MRI scanning of the midbrain was normally finished within 30 min. Animals were taken out of the cradle and given atipamezole (0.5 mg/kg im) to reverse the effects of the medetomidine. They were then continuously

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*Fig. 2. Assessment of midbrain damage. A: an example of a section through the inferior colliculus (IC). The central nucleus of the inferior colliculus (ICc) was more darkly stained than the surrounding areas (dashed line). B: an example of a section through the superior colliculus (SC). The superficial layers of SC are marked by a boundary (arrow) that can be recognized under the microscope. C: sketches showing examples of midbrains from 1 normal and 2 lesioned animals. Darkened areas show the residual, postlesion IC (top) and superficial layers of the SC (bottom). The animal with the smaller lesion (middle) has some residual SC and IC bilaterally, whereas the animal with the larger lesion (right) is missing left SC and IC entirely.*

*Fig. 3. Neuronal response types. A and B: the relative proportions of neuronal response types in AC of all normal (A) and all lesioned ferrets (B). A, auditory neurons; M, multisensory neurons; V, visual neurons. C–F: representative poststimulus response histograms for the different neuronal response types in cross-modal AC. A in top traces indicates the time course of the auditory stimulus, OX indicates the time of optic chiasm stimulation, and V indicates the timing of the light stimulation. C: auditory neurons respond to sound but not optic chiasm stimulation. D: multisensory neurons respond to both sound and optic chiasm stimulation. E: this visual neuron responded to optic chiasm stimulation but not sound. F: this visual neuron was also responsive to light.*

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*J Neurophysiol • VOL 105 • APRIL 2011 • www.jn.org*
monitored over the next 1–2 h before being returned to the colony to ensure that they were completely recovered from the drugs.

**Histology.** After electrophysiology, animals were deeply anesthetized with pentobarbital sodium (65 mg/kg) for euthanization and perfusion with phosphate-buffered saline (PBS), followed by 2–4% paraformaldehyde in 0.1 M phosphate buffer (PB). Brains were extracted, postfixed in 4% paraformaldehyde in 0.1 M PB for 24 h, and stored in 30% sucrose in 0.1 M PB at 4°C. After the tissue was infiltrated by the sucrose solution, it was sectioned frozen at 50 μm in the coronal plane for reconstruction of lesions. A series of sections at 200-μm intervals was stained for Nissl substance using cresyl echt violet.

**Analysis.** The size of the residual central nucleus of the inferior colliculus (ICc) and the superficial layers of the superior colliculus (sSC) in each animal’s midbrain was measured from Nissl-stained sections with a Zeiss microscope using Zeiss Axon Vision 3.1 software (Carl Zeiss MicroImaging, Thornwood, NY). The borders of ICc and sSC (areas indicted by the dashed lines in Fig. 2, A and B, and dark areas in Fig. 2C) were very clear on our Nissl-stained sections. The volume of sSC and ICc was calculated as the sum of each of the measured areas multiplied by 200 μm. Proportions of residual midbrain area and volume in the lesioned animals were calculated by comparison with an average midbrain volume derived from five normal animals. Lesioned animals were sorted into small- and large-lesion groups as determined by these measurements (Fig. 2).

**RESULTS**

Twenty-five ferrets in total were used in this study. Ten were entered in the normal group, and 15 received neonatal lesions leading to cross-modal plasticity. Below we characterize and compare the response properties of the 401 AC neurons recorded in the normal group and the 573 AC neurons recorded in the lesioned group.

**Normal AC Contains Primarily Auditory Responses Plus Rare Multisensory Responses**

Normal animals were used in the experiments as a negative control for the effects of the midbrain lesion. Although primary sensory cortices are traditionally defined as brain areas that respond only to a single sensory modality, recent research has challenged this view by reporting the existence of multisensory neurons and neurons responding to other modalities in primary sensory cortices. Bizley et al. (2007) have reported that primary AC in ferrets does contain some auditory/visual bimodal and some visual neurons. To investigate whether and to what extent primary auditory cortices (A1 and AAF) in normal ferrets can respond to visual stimuli under our experimental conditions and methods of analysis, we characterized the response modality of 401 single neurons in AC of 10 normal animals using in vivo extracellular recording. We defined auditory neurons and visual neurons as those that responded to only one modality. Multisensory neurons were defined as those that either responded to both modalities or responded to one modality but were significantly modulated by stimulation from the other modality (criterion of P < 0.05, t-test on number of spikes to single vs. bimodal stimuli, 10 trials or in some cases 15 trials, data obtained from PSTHs) (see Meredith and Stein 1986). We found that 11% of the 401 neurons recorded in AC of normal ferrets were multisensory. These multisensory neurons responded to both sound stimuli and electrical stimulation of the optic chiasm (n = 45, Fig. 3A) but not to stimulation by light. No visual-only neurons were found in our sample of normal animals.

**XMAC Contains Visual, Auditory, and Multisensory Response Types**

We next tested whether auditory responses remain in AC of lesioned animals and whether the ectopic visual inputs to MGN were associated with an increased proportion of multisensory or visual-only units. We predicted that XMAC’s residual inputs from auditory areas would preserve auditory responsiveness, despite earlier reports to the contrary (Roe et al. 1992; Sur et al. 1988). Callosal connections between XMAC and AC in the nonlesioned hemisphere exist (Pallas et al. 1999), and the inferior colliculi are incompletely lesioned in many cases. These inputs could confer auditory responses on XMAC. In support of this prediction, our data showed a high proportion of auditory neurons in XMAC, despite the presence of ectopic visual responses. Multisensory neurons were also found. In all of the lesioned animals considered together, the relative pro-

Table 1. The proportion of neuron types and residual midbrain volumes in lesions animals

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<th>Animal</th>
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<th>Visual, %</th>
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Midbrain volumes for the left (L) and right (R) inferior colliculus (IC) and superior colliculus (SC) were normalized to average midbrain volumes of normal animals.
portion of auditory-only neurons was 56%, the proportion of multisensory neurons was 32%, and the proportion of visual-only neurons (optic chiasm and/or light driven) was 12% (Fig. 3B). Poststimulus time histograms are shown for each response type in Fig. 3, C–F. The existence of auditory neurons in XMAC reveals that the neonatal midbrain lesions and ectopic visual inputs do not eliminate or suppress the auditory function of AC. The presence of a higher than normal proportion of multisensory neurons in XMAC suggests that auditory and visual inputs are more likely to converge in XMAC than in normal AC.

Continuous and Categorical Differences in Response Type Occurred by Altering the Extent of Midbrain Sparing

The above-described finding that auditory and visual responses can be made to coexist in XMAC allowed us to address the relationship between the two response types in more detail, and in particular to examine how the induction of visually responsive areas in AC would affect normal auditory processing. In patients with a sensory deficit or damage that deafferents a brain area, invasion of cross-modal inputs often occurs to varying extents and at some point may become maladaptive. We wanted to determine whether progressively increasing the extent of visual invasion of XMAC would result in competition (intersensory suppression) or cooperation (multisensory convergence) between modalities. Given previous reports that auditory responses are absent in XMAC (Roe et al. 1992), we wanted to test whether increasing levels of visual input activity would suppress or eliminate auditory responses. Such a finding could explain why we observed auditory responses in our data set, whereas none were found in the Roe et al. (1992) study in which very large lesions were made.

To investigate the effect that increasing degrees of invasion of ectopic visual inputs would have on auditory responsiveness in AC, we measured the midbrain lesion size in each ferret by using histological techniques and compared this measure with the relative proportions of each response type in a systematic fashion. We quantified midbrain size of the lesioned animals by comparing the volume of the left and right ICc and the left and right sSC in each lesioned animal to that averaged across five normal animals used as a standard of comparison (cf. Fig. 2; Table 1).

Fig. 4. Relationship between midbrain lesion type and size and neuronal response type in AC of experimental animals. A: the proportion of visual neurons was negatively correlated with midbrain size. B: the proportion of visual neurons was negatively correlated with left SC size. C: the proportion of visual neurons was not correlated with left IC size. D: the proportion of auditory neurons was positively correlated with midbrain size. E: the proportion of auditory neurons was positively correlated with left SC size. F: the proportion of auditory neurons was positively correlated with left IC size. Each symbol corresponds to 1 animal (n = 14).
Next, it was necessary to demonstrate that increasing the lesion size would affect the relative proportions of auditory, visual, and multisensory neurons in XMAC. We found that midbrain lesion size was correlated with the proportion of auditory and visual response types in an interesting way. In general, overall lesion size was correlated with the proportion of visual units and inversely correlated with the proportion of auditory units (Fig. 4, A and D).

There was a tighter relationship between spared sSC size and visual responsiveness (r = 0.7, P < 0.006) than between spared ICc size and visual responsiveness (r = 0.48, P = 0.085; compare Fig. 4, B and C). These results suggest that establishment of visual neurons in XMAC relies more on damage to visual midbrain than to auditory midbrain. For auditory responsiveness, the correlations with total midbrain size, sSC size, and ICc size were similar to each other (Fig. 4, D–F). These data show that residual sSC volume predicts the relative proportions of visual and auditory neurons, whereas ICc volume is predictive only of the proportion of auditory neurons, demonstrating that the amount of retinal target area lost (SC lesion) is crucial for determination of neuron types in XMAC when both ICc and sSC are lesioned. It was also notable that even complete ablation of left IC did not eliminate auditory responses in XMAC (Table 1). We did not find any correlation between the proportion of multisensory units and the amount of spared auditory or visual midbrain.

The relationship between lesion size and proportion of auditory units (Fig. 4D) appeared roughly linear, with increasing lesion size correlating with a decreasing proportion of auditory responses. In terms of visual responsiveness, however, as may be predicted by examination of Fig. 4, A and B, there was evidence of an exponentially decreasing relationship (exponential fit: Fig. 4A, r = −0.8, P = 0.0007; Fig. 4B, r = −0.88, P < 0.0001) or perhaps a categorical response to lesion size rather than a progressive, linear response.

XMAC contained light-responsive neurons (that is, neurons that responded to light as well as to optic chiasm stimulation) only in animals in which most of the left midbrain was ablated (~10% residual left midbrain). In animals with >10% residual left midbrain, there were no light-responsive neurons. We used this categorical distinction to divide the cases into a large-lesion or small-lesion group, respectively, and conducted further analyses according to these categories. We performed statistical analysis to examine whether small- vs. large-lesion groups had significantly different residual midbrain sizes. We found that the spared left midbrain size of animals in both the large- (5.4 ± 1.76%, n = 5) and small-lesion groups (43.7 ± 5.34%, n = 10) was significantly smaller than that in the normal group (ANOVA, P < 0.001, Fig. 5). The spared midbrain size in the large-lesion group was also significantly reduced compared with that in the small-lesion group (Tukey’s post hoc test, P < 0.001), allowing us to consider these groups...
along with the nonlesioned group as distinct categories for statistical analyses.

Responsiveness to light requires minimal sparing of visual midbrain. To determine the relationship between lesion group membership and distribution of response types, we calculated the relative proportions of auditory, visual, and multisensory responders in each group. This analysis allowed us to compare the proportion of neuronal responses among groups, including the proportion of multisensory neurons that had not been shown in the correlation analysis.

Animals in the small-lesion group had few visual neurons in XMAC (1.8 ± 0.96%, n = 8 of 414 neurons from 10 animals), and those neurons responded to electrical stimulation of the optic chiasm but not to light. The low proportion likely results from the minimal redirection of retinal axons to MGN in small-lesion cases (Angelucci et al. 1998). In contrast, 33.8 ± 7.92% of recorded neurons in the large-lesion group were visual neurons (n = 61 of 159 neurons from 5 animals, Fig. 6A). Of the 61 visual neurons, 48 (78.7%) responded to light in addition to optic chiasm stimulation. This is a significant increase (Mann-Whitney U-test, P < 0.003) and represents a categorical difference between the large- and small-lesion groups. These results indicate that considerable visual information was reaching AC in the large-lesion animals.

Auditory neurons become visual rather than converting to multisensory neurons. Although auditory neurons were found in all groups, the proportion of auditory neurons to total recorded neurons in both groups of lesioned animals varied with lesion size (large-lesion group: 43.8 ± 6.47%, n = 65 of 159; small-lesion group: 60.0 ± 3.22%, n = 256 of 414, normal group: 88.6 ± 1.65%, n = 356 of 401; ANOVA, P < 0.001, Fig. 6B). Furthermore, the proportion of auditory neurons in the large-lesion group was less than that in the small-lesion group (P = 0.003, Tukey’s post hoc test). These results suggest that increasing lesion size resulted in an increase in visual neurons largely at the expense of auditory neurons, rather than a conversion of auditory neurons into multisensory neurons.

Multisensory neurons were found in all groups. We found that neurons in AC of all three groups responded to both auditory and optic chiasm stimulation (multisensory neurons in the large-lesion group: mean 22.4 ± 5.60%, n = 33 of 159; small-lesion group: mean 38.3 ± 3.51%, n = 150 of 414, normal group: mean 11.5 ± 1.65%, n = 45 of 401; ANOVA, P < 0.001, Fig. 6C) but, interestingly, that the proportion of multisensory neurons in the small-lesion group was significantly higher than the proportion of multisensory neurons in the normal group (Tukey’s post hoc test, P < 0.001) and in the large-lesion group (Tukey’s post hoc test, P < 0.05). Although the large-lesion group contained more multisensory neurons than did the normal group, statistical analysis indicated that there was no significant difference (large-lesion vs. normal, P = 0.11). This probably results from the high proportion of purely visual neurons in the large-lesion group and suggests that the response of XMAC to minimal invasion of visual activity (making multisensory neurons) is different from its response to substantial visual influence (making purely visual neurons). This result has interesting implications for rehabilitation strategies for patients with sensory loss or brain damage that cause cross-modal redirection of afferent inputs. Selective stimulation of the original modality would be expected to produce very different results depending on the amount of redirection.

Fig. 7. Reconstruction of locations of recorded neurons in normal AC. Each panel exhibits data from 1 animal (8 of the 10 cases are shown). Each open circle represents one auditory unit; each shaded circle represents one multisensory unit. Plus signs represent neurons that were responsive to the stimulus, whereas minus signs represent neurons that were not responsive to that modality; thus A+/OX− indicates responsiveness to auditory stimulation but not to optic chiasm stimulation and A+/OX+ indicates responsiveness to both auditory and optic chiasm stimulation. × indicates a nonresponsive site. Scale bar, 1 mm.
Auditory and Multisensory Neurons Have Distinct Spatial Distributions in Normal Animals

To determine whether neuronal responses to different modalities are preferentially located in different regions of AC, we calculated the distribution of recorded neurons in the normal AC across the four quadrants (cf. Fig. 2). Figure 7 shows examples of the penetration locations and neuronal response types in recordings of normal AC from 8 of the 10 normal animals. We found that auditory neurons were distributed randomly across the entire AC, but most multisensory neurons were located laterally. Using statistical analysis, we found that auditory neurons were evenly distributed ($\chi^2$, $P > 0.05$). However, the distribution of multisensory neurons was skewed to one of the four quadrants ($\chi^2$, $P < 0.001$, analysis of residuals: $R = 6.93 > R_{0.001} = 3.29$) (Fig. 8). The polar plot (Fig. 8A) shows that the population of multisensory neurons was preferentially located in the lateroposterior quadrant (Q4; see Fig. 2) in normal animals. The observed incidence of multisensory neurons in Q4 was significantly higher than the expected value of 25% (Fig. 8B). These data show that auditory responses in normal animals are distributed evenly across the AC, whereas multisensory neurons are preferentially located lateroposteriorly.

The Spatial Distribution of Auditory, Visual, and Multisensory Neurons in Lesioned Animals Was Different From Normal

Spatial distribution of neuronal response types in small-lesion cases. We next investigated whether neurons with auditory and visual responses in AC of small-lesion animals would be segregated as seen for multisensory neurons in normal AC. Such clustering would likely facilitate efficient processing of visual information separately from auditory information after the midbrain injury. Eight examples of raw data are presented in Fig. 9 to indicate recording locations and neuronal response types in each small-lesion animal. Pooled data from all 10 small-lesion animals showed that auditory neurons in small-lesion AC were randomly distributed across the four quadrants ($\chi^2$, $P > 0.05$, Fig. 10). Multisensory neurons could also be found in any of the quadrants in XMAC of small-lesion animals but were more likely to be found in Q4. The number of multisensory neurons in the anteromedial quadrant (Q1) was below the expected value for a random distribution ($\chi^2$, $P < 0.01$, analysis of residuals: $R = 2.79 > R_{0.01} = 2.58$, Fig. 10B), and the number in the lateroposterior quadrant (Q4) was above the expected value ($\chi^2$, $P < 0.001$, analysis of residuals: $R = 3.61 > R_{0.001} = 3.29$, Fig. 10B). In addition to auditory and multisensory neurons, we recorded some purely visual neurons (8 of 414 neurons) that were located exclusively in lateral AC (solid triangles in Fig. 10). These results suggest that ectopic visual inputs can invade the entire AC, even though they are concentrated in the lateral posterior quadrant. This is interesting given the result that Q4 already contains multisensory neurons in normal animals (Fig. 8). Although the multisensory neurons were preferentially located in the lateroposterior quadrant of both normal and small-lesion animals, the extent of clustering in small-lesion animals was reduced compared with that in normal animals. These findings imply that the random distribution of ectopic visual inputs to XMAC may weaken the tendency of visually responsive neurons in normal AC to cluster.

In addition to analyzing the distribution of neuronal responses in the four quadrants, we measured the distance between each single unit and its neighbors to further test the segregation hypothesis. We found that the average distance between pairs of sound-responsive neurons was significantly shorter than the average distance between auditory and multisensory neurons (ANOVA, $P < 0.05$, Fisher LSD method, Fig. 10C). This finding suggests that auditory neurons are more likely to cluster with each other than with multisensory neurons. This result may be attributable to the smaller number of multisensory responses compared with auditory responses in XMAC of small-lesion animals. We were prevented from including visual responses in the analysis because some animals had only one visual neuron in the entire AC.
Spatial distribution of neuronal response types in large-lesion cases. Because some response type clustering was seen in AC of small-lesion cases, we wondered whether the more extensive visual inputs resulting from larger lesions would either promote or reduce clustering of neuronal response types in XMAC. Note that it is difficult to generate animals with large midbrain lesions, thus there is a smaller number \((n = 5)\) of large-lesion cases compared with small-lesion cases (Fig. 11). We found that auditory, visual, and multisensory neurons were evenly distributed across the four quadrants of AC in large-lesion cases \((\chi^2, P > 0.05, \text{Fig. 12})\). This was different from normal and small-lesion cases, in which the multisensory and visual neurons were preferentially located in the latero-posterior quadrant (Q4). Although there was variation in the sample, there was no significant degree of segregation of neuronal response types in XMAC of the large-lesion group. These results suggest that ectopic visual inputs to AC of large-lesion animals do not segregate.

As with the small-lesion group, we measured the distance between pairs of auditory, visual, and multisensory neurons in the large-lesion group to test the hypothesis that neurons with similar response properties would be clustered together. We compared the average distance between pairs of auditory neurons to the average distance between auditory-visual pairs and auditory-multisensory pairs of neurons. We did not find any significant clustering when we compared average distance between pairs of visual neurons to the average distance between visual-auditory and visual-multisensory pairs (ANOVA, \(P > 0.05\), Fisher LSD method, Fig. 12C). We did find that the average distance between pairs of multisensory neurons was significantly shorter than the average distance between multisensory-auditory pairs (ANOVA, \(P < 0.05\), Fisher LSD method, Fig. 12C) but found no significant difference between pairs of multisensory neurons and between auditory-multisensory pairs. These results suggest that multisensory neurons are closer to each other than to visual neurons. Overall, these results are in agreement with our polar plot data and further suggest that auditory and visual inputs to AC of large-lesion animals do not segregate.

The Latency of Visually Responsive Neurons to Optic Chiasm Stimulation Differed Between Normal and Lesioned Animals

Previous research demonstrated that ectopic visual inputs to XMAC come from the retino-MGN-AC projection (Pallas et al. 1990; Sur et al. 1988), whereas visual inputs to normal AC come from corticocortical projections (Bizley et al. 2007). We found clustering of multisensory neurons lateroposteriorly in normal AC, whereas multisensory neurons were randomly distributed in AC of large-lesion animals. To investigate whether visual inputs to multisensory and visual neurons in lesioned animals came from expanded corticocortical projections or from retino-MGN-AC afferents, we compared the latency of responses to optic chiasm stimulation. The latency to...
optic chiasm stimulation of the multisensory neurons recorded in normal animals \((n = 40)\) was 13.4 ± 1.75 ms, whereas the latency to optic chiasm stimulation of multisensory and visual neurons from lesioned animals was 8.76 ± 1.03 ms \((n = 86)\) in the small-lesion group and 5.72 ± 0.65 ms \((n = 65)\) in the large-lesion group, which was significantly shorter (ANOVA, \(P = 0.015\), small vs. normal; \(P < 0.001\), large vs. normal; Fig. 13). No significant difference was found between small- and large-lesion groups. The comparison between latency of response to optic chiasm stimulation in normal visual cortex and XMAC was reported in a previous study in which the authors showed that visual neurons in XMAC have longer latencies than visual neurons in visual cortex (Roe et al. 1992). Together these results provide further evidence to support the contention that normal AC receives its visual inputs indirectly, perhaps from other cortical areas, but not directly from thalamus, whereas XMAC receives visual inputs more directly, probably from the retina to MGN to AC pathway.

**DISCUSSION**

Early lesions to sensory structures in one sensory modality can result in profound reorganization across multiple sensory pathways due to the many interconnections between structures (Karlen et al. 2006; Kingsbury et al. 2000, 2002; Pallas et al. 1990; Pallas and Sur 1993). The fact that such reorganization can result in cross-modal connections has important clinical implications for recovery from perinatal brain damage, because the different modalities could either cooperate or compete with each other. We found that auditory neurons and multisensory neurons coexist with visual neurons in XMAC after recovery from neonatal midbrain damage. The existence of multisensory neurons indicates that the two modalities can converge and cooperate to activate individual target neurons, rather than competitive suppression of one input by the other. The proportions of auditory and visual neurons were directly related to the amount of residual midbrain tissue. Rather than being segregated from visual neurons, auditory neurons were evenly distributed across XMAC of lesioned animals, and an increase in the number and a broadening of the distribution of multisensory neurons was observed. These findings are reminiscent of phenomena such as acquired auditory-tactile synesthesia, which was reported in a patient following recovery from a thalamic infarct. In this patient, sound induced blood oxygen level-dependent (BOLD) responses in somatosensory cortices (Beauchamp and Ro 2008; Ro et al. 2007). Collectively, these results suggest that both cooperation and competition between the two input modalities are involved in the reorganization of sensory areas after damage to sensory inputs.

![Figure 10](http://jn.physiology.org/)

**Fig. 10.** Distribution of neuronal response types in AC across the population of small-lesion cases. A: polar plot of pooled data from all 10 animals. Open circles represent auditory neurons, shaded circles or triangles represent multisensory neurons, and solid triangles represent visual neurons. B: the proportion of neurons in each quadrant (pooled data). The distribution of auditory neurons was even across quadrants \((\chi^2; \ P > 0.05)\), but the numbers of multisensory neurons in Q1 and Q4 were significantly different from expected values \((\chi^2; **P < 0.01; ***P < 0.001)\), with the numbers significantly higher in Q4 and lower in Q1. Visual neurons were located only in Q3 and Q4. The proportion of visual neurons in Q4 was significantly higher than expected \((\chi^2; **P < 0.01)\). The dashed line at 25% indicates the value expected if response types were evenly distributed. C: the average distance between single units of each response type in AC. A–A is the average distance between all pairs of auditory neurons, A–M is the average distance between all auditory and multisensory pairs of neurons, and M–M is the average distance between all pairs of multisensory neurons. Each symbol represents the mean of average distances for each comparison type from 1 animal. The average distance between auditory neurons was less than that between multisensory neurons or between auditory and multisensory neurons. \(*P < 0.05\) (ANOVA).
Auditory Function is Retained in Core Auditory Cortical Regions Despite Visual Inputs

Previous studies using neonatal midbrain lesions in ferret kits focused on visual responses in XMAC (Roe et al. 1990, 1992; Sur et al. 1988; von Melchner et al. 2000). Those studies, which used very large midbrain lesions, reported that there was no residual auditory function in cross-modal AC. We used lesions of varying size and found that considerable auditory function is retained despite the visual input, especially in animals with smaller lesions. This fortuitous finding allowed us to examine the effect of different degrees of cross-modal input on development and plasticity of AC in response to deafferentation.

Our finding that a large proportion of the neurons in XMAC retain auditory responsiveness supports the hypothesis that residual auditory afferents can compensate even for complete loss of the ipsilesional inferior colliculus but raises the question of where the auditory information derives from. The most likely sources of auditory input to XMAC are the ipsilateral MGN [as a conduit of input from spared contralateral ICc (Angelucci et al. 1998; Moore et al. 1998)] and the contralateral AC [via the corpus callosum (Pallas et al. 1999)] (Fig. 14). Whether the auditory neurons in XMAC function as they would in normal animals is an important question and is the subject of a current study. Preliminary results suggest that function is somewhat compromised (Mao and Pallas 2010).

Competition Between Visual and Auditory Inputs May Determine Response Type

Our results show that with decreasing size of IC and SC, the percentage of recorded neurons responding to sound went down and the percentage of visual neurons went up. These results are consistent with competition as an explanatory mechanism (Crair et al. 1997; Hubel and Wiesel 1962; Stryker 1982; Stryker and Harris 1986). During recovery, when the normal sensory drive may be maximally compromised, activity-dependent processes may allow invading, cross-modal inputs to outcompete preserved inputs from the normal pathway. These findings suggest that optimizing rehabilitation of patients suffering from sensory dysfunction or brain damage will require not only increasing the activity of the original inputs but also decreasing activity in the ectopic inputs. For example, rearrangement of somatosensory circuits in early blind humans can degrade somatosensory representations (see Sathian and Stillia 2010 for review; Sterr et al. 2003), and cross-modal changes in deaf patients can interfere with the success of cochlear implants (Lee et al. 2001).

Our findings may also be of relevance to studies on recovery from partial deafness following cochlear damage. Fallon et al. (2009), using neonatally deafened cats, found that large portions of A1 were nonresponsive to sound activation of cochlear implants. These nonresponsive regions may actually be visually responsive, perhaps interfering with the efficacy of cochlear implants due to loss of territory for sound processing. Similar loss of auditory cortical territory for sound processing may result from damage to IC due to disease or injury (Bognar et al. 1994; Hoistad and Hain 2003; Kimiskidis et al. 2004; Lee et al. 2009; Masuda et al. 2000; Meyer et al. 1996; Musiek et al. 2004). Understanding how to manipulate competition between sensory modalities converging on a cortical territory would be helpful in designing clinical therapies.

Unisensory Auditory or Visual Neurons are Intermingled Within Cross-Modal AC

In contrast to research on cortical plasticity within one modality, this study addressed how ectopic, cross-modal inputs that invade a deafferented cortex affect normal function. We examined the possibility that both modalities could function independently through the segregation of their representations in XMAC. Neurons with similar response properties tend to be clustered together in unisensory sensory cortex (Hubel and Wiesel 1962, 1963; see Mountcastle 1997 for review) and in
multisensory cortex (Dahl et al. 2009). Activity-dependent sorting of inputs can drive spatial segregation of different response types (Miller et al. 1989; Reh and Constantine-Paton 1985). As in other mammals, primary auditory cortex in ferrets maps sound frequency in one dimension (Kelly et al. 1986; Phillips et al. 1988), whereas primary visual cortex maps visual space in two dimensions (Law et al. 1988). If this remains the case in XMAC, it seems unlikely that visual and auditory neurons would be simultaneously active. It has been suggested that evolutionary pressure causes neurons with similar response properties to group together to reduce axon length and connection distance (Chklovskii and Koulakov 2004; Chklovskii et al. 2002; Kaas 2006; Ringo 1991). Such a tendency would also reduce the difficulty of wiring developing circuits appropriately. We reasoned that this economic pressure along with activity-dependent sorting could induce clustering of neurons with similar responses on an acute basis after brain damage and reorganization. For this reason, and because we found previously that callosal, auditory connections between the nonlesioned hemisphere and the ipsilesional AC were shifted laterally in AC (Pallas et al. 1999), we expected that neurons with auditory responses in XMAC would be segregated from those with visual responses. Instead, visual and auditory responses were intermingled throughout the entire AC. It is interesting that the competitive interaction between auditory and visual inputs in terms of proportion of response types did not also affect their spatial distribution within AC. It is possible that microclusters of similarly responding neurons escaped our detection or that processes that cause segregation are not operational in XMAC. At any rate, these results imply that differences in the modality of information carried by the auditory and visual inputs from MGN during postnatal development are not sufficient to induce segregation or splitting of the cortical target areas.

Fig. 12. Distribution of neuronal responses in AC of large-lesion cases. A: polar plot of pooled data from 5 animals. B: the proportion of neurons in each quadrant in relation to total recorded auditory, multisensory or visual neurons in AC. Auditory, multisensory, and visual neurons are randomly distributed across quadrants ($\chi^2$, $P > 0.05$). The dashed line at 25% indicates the value expected if response types were evenly distributed. C: the average distance between pairs of single units of each response type from 1 animal. The average distance between multisensory and visual neurons was significantly greater than that between pairs of multisensory neurons and between pairs of multisensory and auditory neurons. *$P < 0.05$ (ANOVA).

Fig. 13. Latencies of responses to optic chiasm stimulation in normal AC and cross-modal AC. The response latency in AC of the small-lesion group (8.76 ± 1.03 ms) and in AC of the large-lesion group (5.72 ± 0.65 ms) was significantly shorter than that in normal AC (13.4 ± 1.75 ms). No significant difference was found between small- and large-lesion groups. *$P < 0.05$; ***$P < 0.001$ (ANOVA).
Multisensory neurons was seen in AC of large-lesion animals. The extent of clustering is smaller in X-modal AC than auditory processing. The small, darker gray circle in AC represents clustered multisensory neurons in both normal AC and XMAC. Previous studies in ferrets and other species have also reported the existence of multisensory responses in primary AC, and several investigators have thus begun to question the degree of modality specificity in the primary sensory cortices (e.g., Bizley and King 2009; Ghazanfar and Schroeder 2006). Research on primates (de la Mothe et al. 2006; Lakatos et al. 2007; Smiley et al. 2007) and on rodents and carnivores (Bizley et al. 2007; Angelucci et al. 1998; Roe et al. 1993). We propose that the most likely explanation for our data, then, is that new convergences between auditory and visual inputs are made at the level of AC in the lesioned animals.

Although normal AC and XMAC both contain multisensory neurons, our latency data suggest that the origin of visual inputs to these neurons is different in the two cases. The response latency of multisensory neurons to optic chiasm stimulation in XMAC was much shorter than that in normal AC, suggesting that AC in normal animals receives its visual inputs indirectly from other cortical areas but that XMAC receives them more directly. Tracer injections made by Bizley and colleagues (2007) in AC of normal ferrets revealed projections from visual cortical regions to AC that may contribute to the multisensory responses seen there. In XMAC, however, additional visual inputs come from the retina via the MGN (Sur et al. 1988), and these would be expected to exhibit the shorter latencies that we have seen. The connectional differences between multisensory neurons in normal AC and multisensory 2004) has shown that multisensory responses exist in traditionally defined primary sensory cortices (cortices with direct thalamic input, which would include A1 and AAF, defined in this study as AC). In normal ferrets, King and colleagues reported that 15% of recorded units in A1 and AAF had nonauditory inputs, and these were located primarily along the outer edges of AC (Bizley and King 2007; Bizley et al. 2007). We encountered a similar proportion of multisensory to total neurons in normal ferret AC. The multisensory neurons we recorded in the present study were primarily located at the margins of normal AC, but particularly in lateroposterior AC, near the border between A1 and the posterior pseudosylvian and posterior suprasylvian fields. They responded to direct stimulation of the optic chiasm but not to light, suggesting that they receive only weak visual input. Bizley et al. (2007) used a more sensitive method of response analysis that included spike timing information, which may explain why they found greater sensitivity to light. Although we cannot completely rule out the possibility that some visual units that we recorded were located in nonprimary AC, similarly placed recordings using pure tone stimuli indicate to the contrary.

Multisensory Neurons in Normal AC

In addition to auditory neurons, we found multisensory neurons in both normal AC and XMAC. Previous studies in ferrets and other species have also reported the existence of multisensory responses in primary AC, and several investigators have thus begun to question the degree of modality specificity in the primary sensory cortices (e.g., Bizley and King 2009; Ghazanfar and Schroeder 2006). Research on primates (de la Mothe et al. 2006; Lakatos et al. 2007; Smiley et al. 2007) and on rodents and carnivores (Bizley et al. 2007; Campi et al. 2010; Cappe and Barone 2005; Wallace et al. 2004) has shown that multisensory responses exist in traditionally defined primary sensory cortices (cortices with direct thalamic input, which would include A1 and AAF, defined in this study as AC). In normal ferrets, King and colleagues reported that 15% of recorded units in A1 and AAF had nonauditory inputs, and these were located primarily along the outer edges of AC (Bizley and King 2008; Bizley et al. 2007). We encountered a similar proportion of multisensory to total neurons in normal ferret AC. The multisensory neurons we recorded in the present study were primarily located at the margins of normal AC, but particularly in lateroposterior AC, near the border between A1 and the posterior pseudosylvian and posterior suprasylvian fields. They responded to direct stimulation of the optic chiasm but not to light, suggesting that they receive only weak visual input. Bizley et al. (2007) used a more sensitive method of response analysis that included spike timing information, which may explain why they found greater sensitivity to light. Although we cannot completely rule out the possibility that some visual units that we recorded were located in nonprimary AC, similarly placed recordings using pure tone stimuli indicate to the contrary.

Multisensory Neurons in XMAC

In XMAC the proportion of multisensory neurons was much higher than in normal AC, but it was lower in large-lesion animals than in small-lesion animals. This was contrary to expectation, because the large-lesion group had more visual input to AC than the small-lesion group, leading to more potential interaction between auditory and visual afferents. One possible explanation is that sensory cortical neurons are more likely to be multisensory when they have weaker cross-modal inputs, perhaps because stronger inputs would outcompete and displace the original modality. Another possibility is that multimodal responsiveness represents an intermediate state between an auditorily dominated normal AC and a visually dominated XMAC in cases with large lesions.

It is possible that multisensory responses are first created in MGN before reaching XMAC, although previous investigation of retino-MGN projections reported that they were clustered and segregated in small subregions of the ventral MGN, arguing against convergence at the thalamic level (Angelucci et al. 1997; Angelucci et al. 1998; Roe et al. 1993). We propose that the most likely explanation for our data, then, is that new convergences between auditory and visual inputs are made at the level of AC in the lesioned animals.

Although normal AC and XMAC both contain multisensory neurons, our latency data suggest that the origin of visual inputs to these neurons is different in the two cases. The response latency of multisensory neurons to optic chiasm stimulation in XMAC was much shorter than that in normal AC, suggesting that AC in normal animals receives its visual inputs indirectly from other cortical areas but that XMAC receives them more directly. Tracer injections made by Bizley and colleagues (2007) in AC of normal ferrets revealed projections from visual cortical regions to AC that may contribute to the multisensory responses seen there. In XMAC, however, additional visual inputs come from the retina via the MGN (Sur et al. 1988), and these would be expected to exhibit the shorter latencies that we have seen. The connectional differences between multisensory neurons in normal AC and multisensory...
neurons in AC of lesioned animals are likely to account for the latency difference and may result in different response properties as well.

The expanded proportion of multisensory neurons in XMAC is intriguing. Previous perceptual studies on lesioned ferrets with cross-modal visual input to AC argued that they could “see” rather than “hear” visual cues in the rewired AC (von Melchner et al. 2000). Recent clinical studies showing that thalamic lesions can produce synesthesia (Beauchamp and Ro 2008; Ro et al. 2007) imply that subjects may not be able to identify sensory modalities accurately after cross-modal plasticity, however.

In conclusion, the data from this study provide information about the recovery of sensory function after damage to afferent pathways. They suggest a mechanism whereby visual takeover of auditory cortex during cross-modal plasticity might interfere with auditory function through competition for cortical territory. Appropriate and inappropriate inputs can coexist in the affected cortex without strict segregation, and this may not only interfere with efficient processing, but also create barriers to rehabilitation of the compromised modality. Understanding the processes leading to the coexistence of neurons with different functional roles in the affected cortical areas would be important for designing effective rehabilitation strategies for patients during recovery.

ACKNOWLEDGMENTS

We thank Dr. Xiaoping Hu’s group at Emory University for conducting the MRI scans, Dr. Yu-sheng Hsu’s laboratory at Georgia State University (GSU) for advice on statistics, Taylor Brooks for help with data analysis, and both Dr. Jeff Ko at Purdue University and Dr. Andrew King’s group at University of Oxford for advice on ferret anesthesia. Drs. Charles Derby, Donald Edwards, and Robert Liu provided advice on experimental design, and members of the Pallas laboratory gave helpful comments on the manuscript as well as technical assistance. We are indebted to the Department of Animal Resources at GSU, and particularly Cindy Marshall, for their dedication to animal welfare and expert animal care.

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No conflicts of interest, financial or otherwise, are declared by the author(s).

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