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PROJECTIONS FROM AUDITORY CORTEX CONTACT ASCENDING PATHWAYS THAT ORIGINATE IN THE SUPERIOR OLIVE AND INFERIOR COLLICULUS

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Abstract

The superior olivary complex (SOC) and inferior colliculus (IC) are targets of cortical projections as well as sources of major ascending auditory pathways. This study examines whether the cortical projections contact cells in the SOC or IC that project to higher levels. First, we placed an anterograde tracer into the auditory cortex to label cortico-olivary axons and a retrograde tracer into the IC to label olivocollicular cells in guinea pigs. Cortical axons contacted many labeled cells in the ipsilateral SOC and fewer labeled cells in the contralateral SOC. Contacted cells projected to the ipsilateral or contralateral IC.

In a second experiment, we labeled corticocollicular axons with an anterograde tracer and injected retrograde tracers into the medial geniculate (MG) to label colliculogeniculate cells. In the IC ipsilateral to the cortical injection, many cortical axons contacted colliculogeniculate cells in the dorsal cortex and external cortex of the IC. The contacted cells projected to the ipsilateral MG or, less often, to the contralateral MG.

The results indicate that cortical projections are likely to contact cells in the SOC and IC that project to higher centers. This suggests that auditory cortex can modulate the ascending auditory pathways at multiple levels of the brainstem.

Keywords

medial geniculate; corticofugal; feedback loops; efferent system; descending pathways

1. INTRODUCTION

Auditory cortex is a source of descending projections to auditory nuclei in the diencephalon, midbrain, pons and medulla. These projections have been conceptualized as forming feedback loops and descending chains (Huffman and Henson, 1990; Spangler and Warr, 1991; Winer et al., 1998). Feedback loops comprise cortical projections to subcortical nuclei that project back to cortex, directly or indirectly, allowing cortex to control its inputs from lower centers. One

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well-documented loop is formed by cortical projections to thalamocortical cells in the medial geniculate nucleus (MG) (Bartlett et al., 2000). In this loop, cortical axons make synaptic contact with cells that project directly back to cortex.

Other loops involve cortical projections to cells located at earlier stages of the ascending auditory pathways. We have provided evidence that cortical projections to the cochlear nucleus contact cells that project to the inferior colliculus (IC) (Schofield and Coomes, 2005b). Cortical projections to the superior olivary complex (SOC) may also participate in loops. The cortical projections terminate in the periolivary nuclei as well as the lateral superior olivary nucleus (Coomes and Schofield, 2004a), both of which are major sources of projections to the IC (Schofield and Cant, 1992). However, whether cortical axons contact olivary cells that project to the IC is unknown.

Cortical projections to the IC may also participate in loops. The IC is a major target of cortical projections and is the primary source of ascending auditory projections to the MG. Mitani et al. (1983) recorded intracellularly from colliculogeniculate cells identified by antidromic stimulation of the MG in cats. These cells showed short-latency excitatory postsynaptic potentials (as well as longer latency excitatory and inhibitory postsynaptic potentials) following electrical stimulation of the ipsilateral auditory cortex. The authors suggested that the short-latency excitation resulted from direct cortical inputs to the colliculogeniculate cells. These inputs have yet to be demonstrated anatomically, and questions remain as to where in the IC such contacts might occur.

For the present study, we combined anterograde labeling of corticofugal axons with retrograde labeling of brainstem cells that give rise to ascending projections. In the first experiment, we injected one fluorescent tracer into auditory cortex to label corticofugal axons and different fluorescent tracers into one or both inferior colliculi to label olivocollicular cells. We then examined the SOC for contacts between the labeled axons and the labeled cells. In the second experiment, we labeled cortical axons as before and injected fluorescent tracers into one or both medial geniculate nuclei to label colliculogeniculate cells. We then examined the IC for evidence of contacts between the cortical axons and the colliculogeniculate cells.

2. MATERIALS AND METHODS

Experiments were performed on adult albino guinea pigs (270 – 690 g; either sex) obtained from Charles River Laboratories, Inc. (Wilmington, MA). All procedures were approved by the Institutional Animal Care and Use Committee and administered following the National Institute of Health guidelines for the care and use of laboratory animals.

2.1. Surgery and perfusion

Prior to surgery, each guinea pig was anesthetized with halothane (3.5% for induction, 2.5–2.75% for maintenance) in a mixture of oxygen and nitrous oxide. The guinea pig was premedicated with atropine sulfate (0.08 mg/kg, i.m.) to reduce bronchial secretions, and an antibiotic ointment (Neosporin Ophthalmic) was placed in each eye to prevent drying of the cornea during anesthesia. The animal was then placed in a stereotaxic holder on a feedback-controlled heating pad to maintain body temperature.

During surgery, an incision was made in the scalp and the margins of the incision were injected with a long-lasting local anesthetic (0.25% bupivacaine; Sensorcaine; Astra USA, Inc., Westborough, MA). Stereotaxic coordinates were used to guide all injections. A dental drill was used to open the skull at appropriate locations.

Injections into cortex—The skull was opened over the temporal lobe. Fluorescein dextran (FD, 10% solution in saline; molecular weight = 10,000; Molecular Probes, Inc., Eugene OR) or FluoroRuby (FR, 10% solution in saline; molecular weight = 10,000; Molecular Probes) was injected at 6–56 sites with a 10 μ l Hamilton syringe (Table 1). A volume of 0.1 – 0.2 μ l was injected at each site, yielding a total injection volume of 0.9 – 6.4 μ l.

Injections into the MG—Fast Blue (FB, 5% aqueous solution; Sigma) was injected into one MG in 7 cases and FR was injected into the opposite MG in 4 cases (Table 1). In the first 3 cases, FB was injected with a micropipette (25–28 μ m inside diameter) attached to a Nanoliter Injector (World Precision Instruments, Sarasota, FL). A total of 0.23 μ l (10 pulses, 23 nl/pulse; 1 minute between pulses) was injected at a single site in the MG. In the other 4 cases, the tracers were injected into each MG using 10 μ l Hamilton syringes. A different syringe was used for each tracer. On each side of the brain, a volume of 0.15–0.2 μ l was injected at each of 4 sites in the thalamus.

Injections into the IC—Eight animals received injections into one or both IC (Table 1). Tracers included FD, FR and FB as described above, as well as FluoroGold (FG; 4% aqueous solution; FluoroChrome, Inc., Englewood, CO). Each tracer was injected with a different 10 μ l Hamilton syringe. For each IC injection, tracer was deposited at 4 sites (0.2 μ l/site).

Following the injections, the exposed brain was covered with Gelfoam and the scalp was sutured. To provide 48 hours of post-operative analgesia, Ketoprofen (3 mg/kg, i.p.) was injected immediately after surgery and again a day later. If the animal was to survive more than ten days, it was re-anesthetized and the sutures were removed on the tenth post-operative day.

After 5 to 15 days, the animal was sacrificed with CO₂ gas (inhalation, 10 minutes) or overdose of sodium pentobarbital (440 mg/kg, i.p.), and the animal was perfused through the aorta with approximately 100 ml of Tyrode's solution (pH 7.4), 350 ml of 4% paraformaldehyde in 0.1 M phosphate buffer, pH 7.4 (PB), and 350 ml of 4% paraformaldehyde/10% sucrose in PB. The brain was then removed and stored at 4°C in 4% paraformaldehyde/30% sucrose in PB. On the following day the injected cortex was photographed for reconstruction of the cortical injection site (described below). The cortex was then separated from the brainstem and thalamus. The cortex was frozen and cut in the transverse plane on a sliding microtome. The brainstem (with thalamus) was split at the midline and each half was frozen and cut in the parasagittal plane. All pieces of tissue were cut on a sliding microtome into 40, 50, or 80 μ m thick sections. Sections were collected in six series, at least four of which were mounted on gelatin-coated slides and allowed to dry. One series was counterstained with thionin for the identification of cytoarchitectonic borders. All sections were coverslipped with DPX (Aldrich Chemical Company, Inc., Milwaukee, WI).

2.2. Data analysis

Surface photographs of cortical injections were taken with both natural light and fluorescence filtering using a digital camera (MagnaFire, Optronics, Inc.; Goleta, CA). The images were then related to physiological maps of auditory cortex in the guinea pig (Wallace et al., 2000, 2002) using reconstruction techniques described previously (Coomes and Schofield, 2004a). Transverse sections through cortex were also evaluated to assess injections relative to cortical layers. Cortical layers were identified in adjacent thionin-stained sections according to criteria described previously (Schofield et al., 2006).

Injections into the MG or IC were drawn with a NeuroLucida system (MicroBrightField, Williston, VT) attached to a Zeiss Axioplan 2 fluorescence microscope. Adjacent sections stained with thionin were used to draw nuclear borders. The MG was identified as described by Redies et al. (1989). The subdivisions of the IC - central nucleus, dorsal cortex (ICd), and

external cortex (ICx) - were identified using criteria described in guinea pigs and rats (Faye-Lund and Osen, 1985; Malmierca et al., 1995). Nuclei within the SOC were distinguished according to criteria described previously (Schofield and Cant, 1991, Coomes and Schofield, 2004a).

At least every 6th section through the IC and SOC was examined with a 40x lens or 63x oil immersion lens on the Zeiss Axioplan 2 microscope. Cortical boutons were identified as swellings along the course or at the end of an axon. Boutons that appeared to contact retrogradely-labeled cells were photographed with an Optronics Magnafire camera attached to the microscope. We will refer to these apparent contacts simply as “contacts”; of course, conclusions regarding synapses will have to be verified with subsequent electron microscopic studies. Adobe Photoshop was used to adjust brightness, contrast, and color balance, to arrange and label photomicrographs, and to overlay images collected with different fluorescence filters.

3. RESULTS

We did two series of experiments. The first focused on cortical inputs to olivocollicular cells and the second on cortical inputs to colliculogeniculate cells. We first describe the cortical injections, which were similar in both experiments.

3.1 Cortical injections

Figure 1a shows a representative injection site in temporal cortex. The injections generally spread through most of the cortical layers. Some of the injections spread into white matter (not shown). Cases with these injections showed patterns of terminal labeling similar to cases in which the injections were restricted to layers I–VI. Examination of the thalamus, where retrograde labeling was concentrated in the MG, confirmed that the cortical injections were centered in auditory cortex. The injections typically spread across temporal cortex to include multiple auditory areas (Fig. 1b). According to the maps described by Wallace et al. (2000; 2002), the injections generally included both core and belt areas of auditory cortex. The injection in the case illustrated appeared to spread into some surrounding areas that are not well-defined. In another case (GP369), the injection appeared to encroach on somatosensory cortex. The results in this case were similar to those in other cases. Several animals had smaller injections that appeared to be confined to a few (2–3) cortical areas. These smaller injections always encompassed a large portion of primary auditory cortex (A1) or the dorsocaudal field (DC). The smaller injections labeled fewer corticofugal axons, but the results were similar qualitatively to those from other cases.

3.2. Experiments to identify cortical contacts on olivocollicular cells

Figure 2 shows two representative injection sites in the IC. All injections centered on the central nucleus and included parts of the dorsal and external cortices. None of the injections spread into the contralateral IC or into adjacent structures. The IC injections labeled many cells in numerous auditory areas, including the cochlear nuclei, SOC, nuclei of the lateral lemniscus and auditory cortex. In most areas, the retrograde label was limited to the cell body and proximal dendrites. Within the SOC, cells were observed in a pattern described previously (Schofield and Cant, 1992). Briefly, cells were located in the medial and lateral superior olivary nuclei and in all periolivary nuclei except the posteroventral periolivary nucleus. Many more cells were labeled on the ipsilateral side than on the contralateral side.

Cortico-olivary axons were distributed in a pattern described previously (Coomes and Schofield, 2004a). Axons and boutons were more numerous on the ipsilateral side than on the contralateral side. On both sides, the majority of boutons were located in the ventral nucleus of the trapezoid body and the superior paraolivary nucleus. Additional boutons were present

in the lateral superior olivary nucleus and in the remaining periolivary nuclei. We examined each area that contained labeled axons for contacts with labeled cells.

The majority of labeled boutons were located in the neuropil, among labeled cells but not in obvious contact. The retrograde tracers rarely labeled the dendrites beyond the first branch point, so it was impossible to determine whether contacts occur on distal dendrites. However, numerous boutons appeared to contact labeled cell bodies or proximal dendrites. The majority of contacts were observed in the SOC ipsilateral to the cortical injection (Fig. 3). Of these contacts, most were located within the superior paraolivary nucleus and the ventral nucleus of the trapezoid body. A few contacts were also observed in the dorsal periolivary nuclei (dorsal and dorsolateral periolivary nuclei), the lateral periolivary nuclei (anterolateral periolivary nucleus and lateral nucleus of the trapezoid body), and the lateral superior olivary nucleus. Among the cells that were contacted, the majority projected to the ipsilateral IC (Fig 3a–c); fewer projected to the contralateral IC (Fig. 3d, e). No contacts were observed in the medial superior olivary nucleus, the medial nucleus of the trapezoid body or the posteroventral periolivary nucleus.

Cortical projections to the contralateral SOC are much less dense than those to the ipsilateral SOC. Contacts between labeled axons and labeled cells were observed (Fig. 3f–h), but they were far fewer than on the ipsilateral side. Contacts were observed most often in the superior paraolivary nucleus and ventral nucleus of the trapezoid body. Additional contacts were observed in most of the periolivary nuclei and the lateral superior olivary nucleus. Contacts occurred on somas or proximal dendrites of cells that projected to the IC ipsilateral (Fig. 3g, h) or, less often, contralateral (Fig. 3f) to the SOC in which the cell was located.

In summary, cortical axons contacted olivocollicular cells in the ipsilateral SOC much more often than in the contralateral SOC. On each side, more contacts were observed on cells that projected to the ipsilateral IC than onto cells that projected to the contralateral IC.

3.3. Experiments to identify cortical contacts on colliculogeniculate cells

We placed injections into temporal cortex and the MG in seven animals to search for corticocollicular contacts on colliculogeniculate cells. Injections into the temporal cortex were similar to those described above (Fig. 1). Corticocollicular axons were observed bilaterally in the IC with an ipsilateral predominance. On both sides, axons with en passant and terminal boutons were located primarily in the ICx and ICd. On the ipsilateral side, labeled axons and boutons were also scattered sparsely in the ICc.

A representative injection into the thalamus is shown in Figure 4. The injection involved parts of the MG and encroached on some of the surrounding nuclei. In 4 of the 11 thalamic injections, the injection site included parts of the nucleus of the brachium of the IC or posterior thalamic nucleus (not shown). The IC sends axons to these areas (Andersen et al., 1980), so it is possible that some of the labeled IC cells project to these regions instead of the MG. The results in these cases were similar to those in cases with injections confined to the MG. Labeled cells were present bilaterally in each subdivision of the IC. More cells were present in the ipsilateral IC than the contralateral IC.

We examined the IC ipsilateral to the cortical injections for contacts between labeled axons and labeled cells. Numerous contacts were observed in every case, with individual sections often containing multiple examples. The contacts were observed in ICd and ICx; labeled axons and boutons were intermingled in the ICc, but no contacts were observed.

Most of the contacts occurred on cells that projected to the ipsilateral thalamus (Fig. 5a–l). Contacts occurred on both cell bodies and proximal dendrites, and were present in both the

external cortex (Figs. 5a–h) and the dorsal cortex (Fig. 5i–l) of the IC. Although contacts were numerous, the majority of labeled boutons were located in the neuropil, among the labeled cells but not in obvious contact. Whether contacts occur on distal dendrites could not be determined. Fewer contacts were observed on IC cells that project to the contralateral MG (Fig. 5m–o). The contacts were observed in the external cortex and dorsal cortex of the IC.

4. DISCUSSION

The present paper provides evidence for cortical inputs to cells in the SOC and IC that project to higher centers. This is the first evidence that ascending pathways from the SOC may be under direct cortical influence. The results are consistent with physiological studies that suggest direct cortical inputs to IC cells that project to the ipsilateral MG (Mitani et al. 1983). We found additional contacts on IC cells that project to the contralateral MG. In the following discussion, we first consider some technical issues important for interpreting the data. We then discuss functional implications of cortical inputs to ascending brainstem auditory pathways.

4.1. Technical considerations

The most important technical issue is the use of light microscopy to assess the likelihood of synaptic contacts. Our results do not prove that cortical axons contact the labeled olivary or collicular cells, but they do provide evidence strongly suggesting that such contacts exist. Further, they provide important guidance for future physiological or electron microscopic studies by indicating the likely locations of contacts and the projections of the targeted cells. It is noteworthy in this context that some areas contained labeled cortical axons and retrogradely labeled cells, but did *not* have evidence for contacts. This occurred in the SOC (e.g., in the medial nucleus of the trapezoid body) and in the central nucleus of the IC. Our data do not prove that there are no contacts in these areas, but suggest that further studies should be focused on other regions.

Two issues must be addressed with regard to the size of the injection sites. The first issue regards the spread of the injections beyond the intended target. In both experiments cortex was injected with an anterograde tracer. Cortical projections to the IC (Druga et al., 1988; Winer et al., 1998; Bajo and Moore, 2005) and to the SOC (Coomes and Schofield, 2004a) arise from multiple cortical areas. We made large injections into the temporal cortex to maximize the number of labeled axons and thus the likelihood of discovering any cortical targets that might exist. It is probable that all of our cortical injections spread into multiple auditory areas (see discussion in Coomes and Schofield, 2004a). Future experiments, with injections confined to single, physiologically-identified cortical areas, will be needed to assess the contributions of specific cortical areas to the pathways seen in the present study.

In the first experiment, we made large injections of a retrograde tracer into the IC. These injections did not spread beyond the IC borders. The importance of large injections was to maximize the number of labeled cells and thus the chance of observing cortical inputs. Consequently, our injections routinely involved multiple IC subdivisions, including the central nucleus, dorsal cortex and external cortex. Most olivary projections terminate in the central nucleus, but some (e.g., those from the superior paraolivary nucleus) may also terminate in dorsal cortex (González-Hernández et al., 1996). Consequently, we cannot always specify the subdivision targeted by the olivocollicular cells that receive cortical inputs.

In the second experiment, we made large injections of a retrograde tracer into the MG to maximize the number of labeled colliculogeniculate cells. Some injections spread beyond the MG into parts of the nucleus of the brachium of the IC or posterior thalamic nucleus. Because the IC sends axons to these areas (Andersen et al., 1980), IC cells that project to these regions could have been labeled. However, we also observed contacts following smaller injections.

Thus even if some of the labeled cells from the larger injections were not colliculogeniculate cells, the smaller injections confirm our primary conclusion (i.e., cortical axons contact colliculogeniculate cells).

The second issue regarding injection site size concerns incomplete filling of the target. None of our injections are likely to have labeled every cell or axon in the relevant pathways. Of additional importance is the fact that the retrograde tracers labeled only the soma and proximal dendrites of the cells; we would have missed any contacts on distal dendrites or dendritic spines. The distribution of cortical axon terminals on olivary cells is not known, but their distribution on IC cells (in cats, at least) appears to favor dendrites and spines, with relatively few contacts on somas (Jones and Rockel, 1972; Granstrem, 1984). If the same is true in guinea pigs, the limited labeling of spines and distal dendrites means that we may have missed a significant number of contacts. This issue is relevant for both the IC and the SOC, where the majority of labeled boutons were located in neuropil and not clearly associated with a labeled cell. These limitations suggest that cortical inputs to the pathways under study may be far more numerous than we observed.

4.2. Cortical effects on SOC cells

To our knowledge, there have been no physiological studies of cortico-olivary projections. Anatomical studies show that these projections, like those to the IC, originate from cortical layer V cells and therefore likely use an excitatory neurotransmitter such as glutamate (Mulders and Robertson, 2000; Doucet et al., 2002).

The majority of contacts in the SOC occurred on cells in the periolivary nuclei. As a group, the periolivary nuclei are a significant source of projections to the IC (Schofield and Cant, 1992). Unfortunately, very little is known about the physiological properties of olivocollicular cells. In general, olivocollicular cells are interspersed with cells that project to other targets, so physiological data from periolivary nuclei cannot definitely be attributed to olivocollicular cells (Schwartz, 1992). The superior paraolivary nucleus in rats provides an important exception because virtually all the cells in this nucleus project to the IC (Saldaña and Berrebi, 2000). Kulesza et al. (2003) characterized the acoustically-driven responses of single units in the rat superior paraolivary nucleus. They concluded that the cells in this nucleus are important for encoding temporal features of sounds. Presumably, the cortical projections to the superior paraolivary nucleus are involved in regulating temporal coding at this level, but this remains to be tested. The effects of cortical inputs to olivocollicular cells in other olivary nuclei must await further physiological studies.

Numerous studies have demonstrated an important role of inhibition in shaping collicular responses to sound (Kelly and Caspary, 2005). In rats, apparently all the cells in the superior paraolivary nucleus use GABA as a neurotransmitter (Kulesza and Berrebi, 2000). Many additional olivocollicular cells use GABA or glycine as neurotransmitters and are likely to inhibit their targets in the IC (Saint Marie and Baker, 1990; González-Hernández et al., 1996). The present data suggest that some of these inhibitory inputs to the IC are under the influence of the cortex.

4.3. Cortical effects on IC cells

Stimulation of auditory cortex has been shown to have excitatory, inhibitory, or mixed effects on cells in the IC (Mitani et al., 1983; Syka and Popelár, 1984; Goldstein-Daruech et al., 2002; Bledsoe et al., 2003; Tortorolo et al., 1998; reviewed by Casseday et al., 2002; Suga and Ma, 2003). For cells in the ICc, cortical stimulation can affect spontaneous or sound-evoked activity and can alter the cell's selectivity for various parameters of an acoustic stimulus (Mitani et al., 1983; Sun et al., 1996; Yan and Suga, 1996a, b, 1999; Jen et al., 2001; Ma and

Suga, 2001; Jen and Zhou, 2003; Zhou and Jen, 2005; Yan et al., 2005). Fewer studies have examined corticofugal effects on cells in the ICd or ICx (Jen et al., 2002; Syka et al., 1988; Nwabueze-Ogbo et al., 2002; Bledsoe et al., 2003; Popelár et al., 2003). These studies have shown that cortical stimulation produces facilitation and/or suppression of spontaneous or acoustically-evoked activity in ICd and ICx; however, less is known about effects on selectivity for specific stimulus parameters. Additional experiments, similar to those conducted in ICc, are needed to better understand corticofugal effects in these regions.

The study by Mitani et al. (1983) is the only one to use intracellular recording of IC cells to assess the influence of cortical inputs. The results show that cortical stimulation induces both excitatory and inhibitory post-synaptic potentials in ICc cells, consistent with expectations from extracellular recording. The shortest latency responses to cortical stimulation (1.0 – 1.4 msec) were always excitatory, and the authors concluded they were probably monosynaptic. This is consistent with work suggesting that the corticocollicular axons use glutamate as a neurotransmitter (Feliciano and Potashner 1995). Longer latency responses (1.8 – 4.2 msec) included excitatory and inhibitory effects, and are likely to include di- or multisynaptic circuits. Some of these effects are almost certainly due to cortical activation of cells in ICd or ICx that project in turn to the ICc (Jen et al. 2002).

One question raised by comparison of the present work with that of Mitani and colleagues concerns the locations within the IC of contacts between cortical axons and colliculogeniculate cells. Mitani et al. (1983) identified presumptive monosynaptic inputs onto cells in the ICc in cats. We observed many contacts in ICx and ICd, but none in the ICc. There are several possible explanations for this discrepancy. First, contacts in the ICc may have gone unobserved in the present study because of technical limitations (as discussed above). Second, there may be a species difference, with contacts occurring in ICc in cats but not in guinea pigs. It is also quite possible that the ICc has been identified differently in the two species. On the one hand, Mitani et al., described monosynaptic inputs to cells that had narrow frequency tuning and short-latency responses to acoustic stimuli (features usually associated with ICc). On the other hand, the authors stated, by comparison with anatomical studies, that these units occupy a region of the IC that receives dense cortical inputs (a feature generally associated with ICd, not ICc [reviewed by Winer et al., 1998]). Further studies will be needed to resolve this issue.

4.4. Organization of corticofugal projections: loops and chains

Cortical projections have been implicated in descending pathways through both the IC and the SOC. The cortical projections to the SOC contact cells that project to the cochlea (Mulders and Robertson, 2000) or cochlear nucleus (Schofield and Coomes, 2006). Cortical projections to the IC contact cells that project to the ipsilateral or contralateral cochlear nucleus (Schofield and Coomes, 2006). These projections form descending chains that parallel the direct projections to the cochlear nucleus from cortex. Therefore, neural processing in the cochlear nucleus could presumably be affected by direct cortical projections as well as by cortical influences on projections from the IC and the SOC to the cochlear nucleus. The multitude of descending pathways presumably allows for a variety of effects on lower centers (Schofield and Coomes, 2006).

Potential loops from cortex to the IC to the MG and back to cortex have been described previously (e.g., Huffman and Henson, 1990; Spangler and Warr, 1991; Winer et al., 1998). The present study supports the hypothesis of loops through the ICd and ICx to the thalamus. The thalamic projections from these subdivisions terminate primarily in the dorsal and medial subdivisions of the MG, which together project widely to auditory cortex (de Ribaupierre, 1997). These loops could thus affect activity across most or all of auditory cortex and contribute to numerous auditory functions.

Cortical projections to the SOC provide additional opportunities for interaction with ascending pathways. Some of these pathways could form extended loops that project back to cortex on the same side. Axons that originate, for example, from the left auditory cortex terminate on cells in both the left and right SOC that project to the left IC. The left IC provides the major input to the left MG which in turn projects to the left auditory cortex, completing the feedback loop.

On the other hand, axons from the left cortex, for example, can also contact olivary cells that project to the *right* IC. These projections would presumably have their primary influence on the right MG and right cortex. A similarly “crossed” circuit exists for projections from left cortex to left IC with contacts onto cells that project to the *right* MG. These pathways could allow corticofugal projections from one side of cortex to affect the ascending inputs to the opposite cortex. Whether or not one calls these pathways “loops”, it is clear that the corticofugal projections are positioned to influence a large number of ascending auditory pathways.

In conclusion, the auditory cortex is in a position to affect auditory processing at virtually every level of the auditory system. The auditory cortex projects directly and indirectly, via the SOC and IC, to the cochlear nucleus (Schofield and Coomes, 2006). The cortical projections to the SOC and the IC appear to make direct contact with a number of ascending and descending projections from these regions (present study; Mulders and Robertson, 2000; Schofield and Coomes, 2006). At the same time, the SOC and the IC give rise to other projections. For example, many periolivary cells project to other olivary nuclei, such as the medial and lateral superior olivary nuclei (reviewed by Thompson and Schofield, 2000). These projections play important roles in binaural processing; are they also contacted by cortical axons? Cortical projections to the IC appear to contact cells that project to the thalamus (present study) or to the cochlear nucleus (Schofield and Coomes, 2006). Do cortical axons also contact IC cells that project to other targets such as the pons, the superior colliculus, or the contralateral IC? A full understanding of the functions of the corticofugal system awaits answers to these and similar questions.

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List of Abbreviations

A1	primary auditory cortex
AC	auditory cortex
BIC	nucleus of the brachium of the IC
DC	dorsocaudal field of auditory cortex
DCB	dorsocaudal belt of auditory cortex
DRB	dorsorostral belt of auditory cortex

FB	Fast Blue
FD	fluorescein dextran
FG	FluoroGold
FR	FluoroRuby
IC	inferior colliculus
ic	internal capsule
ICc	central nucleus of IC
ICd	dorsal cortex of IC
ICx	external cortex of IC
LGN	lateral geniculate nucleus
MG	medial geniculate nucleus
rs	rhinal sulcus
Rt	reticular nucleus of the thalamus
S	small field of auditory cortex
SC	superior colliculus
SOC	superior olivary complex
Som	somatosensory cortex
T	transition area of auditory cortex
VCB	ventrocaudal belt of auditory cortex

References

- Andersen RA, Roth GL, Aitkin LM, Merzenich MM. The efferent projections of the central nucleus and the pericentral nucleus of the inferior colliculus in the cat. *J Comp Neurol* 1980;194:649–662. [PubMed: 7451687]
- Aschoff A, Ostwald J. Distribution of cochlear efferents and olivo-cochlear neurons in the brainstem of rat and guinea pig. *Exp Brain Res* 1988;71:241–251. [PubMed: 3169161]
- Bajo VM, Moore DR. Descending projections from the auditory cortex to the inferior colliculus in the Gerbil, *Meriones unguiculatus*. *J Comp Neurol* 2005;486:101–116. [PubMed: 15844210]
- Bartlett EL, Stark JM, Guillery RW, Smith PH. Comparison of the fine structure of cortical and collicular terminals in the rat medial geniculate. *Neuroscience* 2000;100:811–828. [PubMed: 11036215]
- Bledsoe SC, Shore SE, Guitton MJ. Spatial representation of corticofugal input in the inferior colliculus: a multicontact silicon probe approach. *Esp Brain Res* 2003;153:530–540.
- Coomes DL, Schofield BR. Projections from the auditory cortex to the superior olivary complex in guinea pigs. *Eur J Neurosci* 2004a;19:2188–2200. [PubMed: 15090045]
- Coomes DL, Schofield BR. Separate projections from the inferior colliculus to the cochlear nucleus and thalamus in guinea pigs. *Hear Res* 2004b;191:67–78. [PubMed: 15109706]
- de Ribaupierre, F. Acoustical information processing in the auditory thalamus and cerebral cortex. In: Ehret, G.; Romand, R., editors. *The Central Auditory System*. Oxford University Press; New York: 1997. p. 317-397.
- Doucet JR, Rose L, Ryugo DK. The cellular origin of corticofugal projections to the superior olivary complex in the rat. *Brain Res* 2002;925:28–41. [PubMed: 11755898]
- Doucet JR, Molavi DL, Ryugo DK. The source of corticocollicular and corticobulbar projections in area Te1 of the rat. *Exp Brain Res* 2003;153:461–6. [PubMed: 13680047]
- Druga, R.; Syka, J.; Rajkowska-Markow, G. Localization of cortical neurons projecting to the inferior colliculus in the rat and guinea pig. In: Syka, J.; Masterton, BR., editors. *Auditory Pathway Structure and Function*. Plenum Press; New York: 1988. p. 293-298.
- Faye-Lund H, Osen KK. Anatomy of the inferior colliculus in rat. *Anat Embryol* 1985;171:1–20. [PubMed: 3985354]
- Feliciano M, Potashner SJ. Evidence for a glutamatergic pathway from the guinea pig auditory cortex to the inferior colliculus. *J Neurochem* 1995;65:1348–57. [PubMed: 7643112]
- Feliciano M, Saldaña E, Mugnaini E. Direct projections from the rat primary auditory neocortex to nucleus sagulum, paralemniscal regions, superior olivary complex and cochlear nuclei. *Auditory Neurosci* 1995;1:287–308.
- Goldstein-Daruech N, Pedemonte M, Inderkum A, Velluti RA. Effects of excitatory amino acid antagonists on the activity of inferior colliculus neurons during sleep and wakefulness. *Hear Res* 2002;389:1–7.
- González-Hernández T, Mantolan-Sarmiento B, Gonzalez-Gonzalez B, Perez-Gonzalez H. Sources of GABAergic input to the inferior colliculus of the rat. *J Comp Neurol* 1996;372:309–326. [PubMed: 8863133]
- Granstrem EE. Ultrastructural organization of AI and AIV projection zones of the cat auditory cortex in the inferior colliculi of the lamina tecti. *Neurosci Behav Physiol* 1984;14:296–302. [PubMed: 6472612]
- Hashikawa T, Kawamura K. Retrograde labeling of ascending and descending neurons in the inferior colliculus. A fluorescent double labeling study in the cat. *Exp Brain Res* 1983;49:457–461. [PubMed: 6641843]
- Huffman RF, Henson OW. The descending auditory pathway and acousticomotor systems: Connections with the inferior colliculus. *Brain Res Rev* 1990;15:295–323. [PubMed: 2289088]
- Jacomme AV, Nodal FR, Bajo VM, Manunta Y, Edeline JM, Babalian A, Rouiller EM. The projection from auditory cortex to cochlear nucleus in guinea pigs: an in vivo anatomical and in vitro electrophysiological study. *Exp Brain Res* 2003;153:467–76. [PubMed: 14504855]
- Jen PH, Sun X, Chen QC. An electrophysiological study of neural pathways for corticofugally inhibited neurons in the central nucleus of the inferior colliculus of the big brown bat, *Eptesicus fuscus*. *Exp Brain Res* 2001;137:292–302. [PubMed: 11355376]

- Jen PH, Zhou X, Zhang J, Chen QC, Sun X. Brief and short-term corticofugal modulation of acoustic signal processing in the bat midbrain. *Hear Res* 2002;168:196–207. [PubMed: 12117521]
- Jen PH, Zhou X. Corticofugal modulation of amplitude domain processing in the midbrain of the big brown bat, *Eptesicus fuscus*. *Hear Res* 2003;184:91–106. [PubMed: 14553907]
- Jones EG, Rockel AJ. Observations on complex vesicles, neurofilamentous hyperplasia and increased electron density during terminal degeneration in the inferior colliculus. *J Comp Neurol* 1972;147:93–118. [PubMed: 4682185]
- Kelly, JB.; Caspary, DM. Pharmacology of the inferior colliculus. In: Winer, JA.; Schreiner, CE., editors. *The Inferior Colliculus*. Springer; New York: 2005. p. 248–281.
- Kulesza RJ Jr, Berrebi AS. Superior paraolivary nucleus of the rat is a GABAergic nucleus. *J Assoc Res Otolaryngol* 2000;1:255–269. [PubMed: 11547806]
- Kulesza RJ Jr, Spirou GA, Berrebi AS. Physiological response properties of neurons in the superior paraolivary nucleus of the rat. *J Neurophysiol* 2003;89:2299–2312. [PubMed: 12612016]
- Ma X, Suga N. Corticofugal modulation of duration-tuned neurons in the midbrain auditory nucleus in bats. *Proc Natl Acad Sci USA* 2001;98:14060–14065. [PubMed: 11707597]
- Malmierca MS, Rees A, Le Beau FE, Bjaalie JG. Laminar organization of frequency-defined local axons within and between the inferior colliculi of the guinea pig. *J Comp Neurol* 1995;357:124–44. [PubMed: 7673462]
- Mitani A, Shimokouchi M, Nomura S. Effects of stimulation of the primary auditory cortex upon colliculogeniculate neurons in the inferior colliculus of the cat. *Neurosci Lett* 1983;42:185–189. [PubMed: 6664630]
- Mulders WH, Robertson D. Evidence for direct cortical innervation of medial olivocochlear neurones in rats. *Hear Res* 2000;144:65–72. [PubMed: 10831866]
- Nwabueze-Ogbo FC, Popelár J, Syka J. Changes in the acoustically evoked activity in the inferior colliculus of the rat after functional ablation of the auditory cortex. *Physiol Res* 2002;51:95–104.
- Perrot X, Ryvlin P, Isnard J, Guenet M, Catenois H, Fischer C, Mauguire F, Collet L. Evidence for corticofugal modulation of peripheral auditory activity in humans. *Cereb Cortex* 2006;16:941–8. [PubMed: 16151174]
- Popelár J, Nwabueze-Ogbo FC, Syka J. Changes in neuronal activity of the inferior colliculus in rat after temporal inactivation of the auditory cortex. *Physiol Res* 2003;52:615–628. [PubMed: 14535838]
- Redies H, Brandner S, Creutzfeldt OD. Anatomy of the auditory thalamocortical system of the guinea pig. *J Comp Neurol* 1989;282:489–511. [PubMed: 2723149]
- Saint Marie RL, Baker RA. Neurotransmitter-specific uptake and retrograde transport of [3H]glycine from the inferior colliculus by ipsilateral projections of the superior olivary complex and nuclei of the lateral lemniscus. *Brain Res* 1990;524:244–253. [PubMed: 1705464]
- Saldaña E, Berrebi AS. Anisotropic organization of the rat superior paraolivary nucleus. *Anat Embryol* 2000;202:265–279. [PubMed: 11000278]
- Schofield BR. Origins of projections from the inferior colliculus to the cochlear nucleus in guinea pigs. *J Comp Neurol* 2001;429:206–20. [PubMed: 11116215]
- Schofield BR. Ascending and descending projections from the superior olivary complex in guinea pigs: different cells project to the cochlear nucleus and the inferior colliculus. *J Comp Neurol* 2002;453:217–225. [PubMed: 12378584]
- Schofield BR, Cant NB. Organization of the periolivary nuclei in pigmented guinea pigs: Distribution of cells that send axon collaterals to the inferior colliculus, the cochlear nucleus, or both. *Assoc Res Otolaryngol Abst* 1991;14:19.
- Schofield BR, Cant NB. Organization of the superior olivary complex in the guinea pig: II. Patterns of projection from the periolivary nuclei to the inferior colliculus. *J Comp Neurol* 1992;317:438–455. [PubMed: 1578006]
- Schofield BR, Coomes DL. Auditory cortical projections to the cochlear nucleus in guinea pigs. *Hear Res* 2005a;199:89–102. [PubMed: 15574303]
- Schofield BR, Coomes DL. Projections from auditory cortex contact cells in the cochlear nucleus that project to the inferior colliculus. *Hear Res* 2005b;206:3–11. [PubMed: 16080994]

- Schofield BR, Coomes DL. Pathways from auditory cortex to the cochlear nucleus in guinea pigs. *Hear Res* 2006;216–217. 81–9. [PubMed: 16597491]
- Schofield BR, Coomes DL, Schofield RM. Cells in auditory cortex that project to the cochlear nucleus in guinea pigs. *J Assoc Res Otolaryngol* 2006;7:95–109. [PubMed: 16557424]
- Schwartz, IR. The superior olivary complex and lateral lemniscal nuclei. In: Webster, DB., et al., editors. *The mammalian auditory pathway: neuroanatomy*. Springer-Verlag; New York: 1992. p. 117–167.
- Spangler, KM.; Warr, WB. The descending auditory system. In: Altschuler, RA.; Bobbin, RP.; Clopton, BM.; Hoffman, DW., editors. *Neurobiology of Hearing: The Central Auditory System*. Raven Press; New York: 1991. p. 27–45.
- Suga N, Ma X. Multiparametric corticofugal modulation and plasticity in the auditory system. *Nature Rev* 2003;4:783–794.
- Sun X, Chen QC, Jen PH. Corticofugal control of central auditory sensitivity in the big brown bat, *Eptesicus fuscus*. *Neurosci Lett* 1996;212:131–4. [PubMed: 8832656]
- Syka J, Popelár J. Inferior colliculus in the rat: neuronal responses to stimulation of the auditory cortex. *Neurosci Lett* 1984;51:235–40. [PubMed: 6514239]
- Syka, J.; Popelár, J.; Druga, R.; Volková, A. Descending central auditory pathway—structure and function. In: Syka, J.; Masterton, BR., editors. *Auditory Pathway Structure and Function*. Plenum Press; New York: 1988. p. 279–292.
- Thompson, AM. Descending connections of the auditory midbrain. In: Winer, JA.; Schreiner, CE., editors. *The inferior colliculus*. Springer; New York: 2005. p. 182–199.
- Tortorolo P, Zurita P, Pedemonte M, Velluti RA. Auditory cortical efferent actions upon inferior colliculus unitary activity in the guinea pig. *Neurosci Lett* 1998;249:172–6. [PubMed: 9682844]
- Wallace MN, Rutkowski RG, Palmer AR. Identification and localization of auditory areas in guinea pig cortex. *Exp Brain Res* 2000;132:445–456. [PubMed: 10912825]
- Wallace MN, Rutkowski RG, Palmer AR. Interconnections of auditory areas in the guinea pig neocortex. *Exp Brain Res* 2002;143:106–119. [PubMed: 11907696]
- Weedman DL, Ryugo DK. Projections from auditory cortex to the cochlear nucleus in rats: synapses on granule cell dendrites. *J Comp Neurol* 1996a;371:311–324. [PubMed: 8835735]
- Weedman DL, Ryugo DK. Pyramidal cells in primary auditory cortex project to cochlear nucleus in rat. *Brain Res* 1996b;706:97–102. [PubMed: 8720496]
- Winer JA, Larue DT, Diehl JJ, Hefti BJ. Auditory cortical projections to the cat inferior colliculus. *J Comp Neurol* 1998;400:147–74. [PubMed: 9766397]
- Yan J, Suga N. Corticofugal modulation of time-domain processing of biosonar information in bats. *Science* 1996a;273:1100–1103. [PubMed: 8688095]
- Yan J, Suga N. The midbrain creates and the thalamus sharpens echo-delay tuning for the cortical representation of target-distance information in the mustached bat. *Hear Res* 1996b;93:102–110. [PubMed: 8735071]
- Yan J, Suga N. Corticofugal amplification of facilitative auditory responses of subcortical combination-sensitive neurons in the mustached bat. *J Neurophysiol* 1999;81:817–824. [PubMed: 10036282]
- Yan J, Zhang Y, Ehret G. Corticofugal shaping of frequency tuning curves in the central nucleus of the inferior colliculus of mice. *J Neurophysiol* 2005;93:71–83. [PubMed: 15331615]
- Zhou X, Jen PH. Corticofugal modulation of directional sensitivity in the midbrain of the big brown bat, *Eptesicus fuscus*. *Hear Res* 2005;203:201–215. [PubMed: 15855045]

Figure 1.

Representative injection site in temporal cortex. **(a)** Photomicrograph of a transverse section showing an injection of fluorescein dextran (dark area) in the left temporal cortex of case GP345. The image was captured in color, converted to grayscale, and the contrast inverted (black to white) to improve visibility. Cortical surface is at the left. Layers are indicated by Roman numerals. wm – white matter. Scale bar = 0.5 mm. **(b)** Surface reconstruction showing the extent of temporal cortex injected with FluoroRuby (black area) in case GP365. Overlaid on the reconstruction are outlines of cortical areas as mapped by Wallace et al. (2000, 2002; see text for details). A1 – primary auditory cortex; DC – dorsocaudal auditory cortex; DCB – dorsocaudal belt; DRB – dorsorostral belt; rs – rhinal sulcus; S – small field; Som –

somatosensory cortex; T – transition area; VCB – ventrocaudal belt; Vis – visual cortex; VRB – ventrorostral belt. Scale bar = 1 mm.

Figure 2.

Fluorescence photomicrographs showing representative injections in parasagittal sections through the inferior colliculus (IC). **(a)** Center of a fluorescein dextran injection site. **(b)** Center of a Fast Blue injection site. Both panels: Case GP376. Rostral is right; dorsal is up. ICc – central nucleus of the IC; ICd – dorsal cortex of the IC; ICx – external cortex of the IC; SC – superior colliculus. Scale bar = 0.5 mm.

Figure 3.

Photomicrographs showing cortical axons in contact with cell bodies (arrows) or dendrites (arrowheads) of olivocollicular cells. Cortical axons were labeled by an injection of FluoroRuby into left auditory cortex. Olivocollicular cells were labeled by injections of fluorescein dextran (green) into the left inferior colliculus (IC) and Fast Blue into the right IC. **(a–c)**. Cortical contacts on cells in the left superior olivary complex (SOC; ipsilateral to the injected cortex) that projected to the left IC. The cells were located in the ventral nucleus of the trapezoid body (a) or the superior paraolivary nucleus (b, c). **(d, e)**. Cortical contacts on cells in the left SOC that projected to the right IC. The cells were located in the lateral superior olivary nucleus (d) or superior paraolivary nucleus (e). **(f)**. Cortical contact on a cell in the right

lateral superior olivary nucleus, contralateral to injected cortex, that projected to the left IC. **(g, h)**. Cortical contacts on cells in the right SOC that projected to the right IC. The cells were located in the ventral nucleus of the trapezoid body (g) or the dorsolateral periolivary nucleus (h). Case GP376. Scale bar = 10 μm .

Figure 4.

Photomicrographs of parasagittal sections showing a representative injection (black) of Fast Blue into the medial geniculate nucleus (MG). Thionin-stained sections through the thalamus were photographed. Adjacent sections were photographed with fluorescence to reveal the injection site. The images were overlaid and the injection sites (black) were drawn on the thionin images. Sections are arranged from lateral to medial (top to bottom), and are spaced 600 μ m apart. Scale bar = 1 mm. BIC – nucleus of the brachium of the inferior colliculus; ic – internal capsule; LGN – lateral geniculate nucleus; Rt – reticular nucleus of the thalamus.

Figure 5.

Photomicrographs showing cortical axons in contact with cell bodies (arrows) or dendrites (arrowheads) of colliculogeniculate cells. Cortical axons were labeled by an injection of fluorescein dextran into left auditory cortex. Colliculogeniculate cells were labeled by injections of FluoroRuby into the left medial geniculate nucleus (MG) or Fast Blue into the right MG. **(a–h)**. Photomicrographs of cortical contacts on cells in the external cortex of the left inferior colliculus that project to the ipsilateral (left) MG. **(i–l)**. Photomicrographs of cortical contacts on cells in the dorsal cortex of the left inferior colliculus that project to the ipsilateral (left) MG. **(m–o)**. Photomicrographs of cortical contacts on cells in the dorsal cortex

of the left inferior colliculus that project to the contralateral (right) MG. Cases GP345 (a, b, d–m); GP348 (c, n, o). Scale bar = 10 μ m.

Table 1

Locations of tracer injections

Case #	Left AC	Right AC	Left MG	Right MG	Left IC	Right IC
GP330	x	FD	x	FB	x	x
GP331	x	FD	x	FB	x	x
GP332	x	FD	x	FB	x	x
GP345	FD	x	FR	FB	x	x
GP346	FD	x	FB	FR	x	x
GP347	FD	x	FR	FB	x	x
GP348	FD	x	FR	FB	x	x
GP365	FR	x	x	x	FD	x
GP367	FR	x	x	x	FD	x
GP368	FR	x	x	x	FD	x
GP369	FD	x	x	x	FG	FR
GP372	FD	x	x	x	x	FR
GP376	FR	x	x	x	FD	FB
GP377	FR	x	x	x	FB	FD
GP380	FR	x	x	x	FD	FB

Abbreviations: AC – auditory cortex; FB – Fast Blue; FD – fluorescein dextran, FG – FluoroGold; FR – FluoroRuby; IC – inferior colliculus; MG – medial geniculate; x – indicates no tracer injection.