Coding for Auditory Space in the Nucleus of the Brachium of the Inferior Colliculus in the Ferret

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Schnupp, Jan W. H. and Andrew J. King. Coding for auditory space in the nucleus of the brachium of the inferior colliculus in the ferret. J. Neurophysiol. 78: 2717–2731, 1997. The nucleus of the brachium of the inferior colliculus (BIN) projects topographically to the deeper layers of the superior colliculus (SC), which contain a two-dimensional map of auditory space. In this study, we have used broadband stimuli presented in the free field to investigate how auditory space is represented in the BIN of the ferret. Response latencies and temporal firing patterns were comparable with those in the SC, and both properties showed some variation with stimulus location. We obtained spatial response profiles at two sound levels (5–15 and 25–35 dB above unit threshold). A large proportion of azimuth profiles (41% in the suprathreshold condition, 80% in the near-threshold condition) presented a single peak, indicating that they were tuned to single regions in space. For some of these units, the preferred speaker position varied considerably with sound level. The remaining units showed predominantly either broad “hemifield” or spatially ambiguous “bilobed” response profiles. At suprathreshold sound levels, the preferred azimuths of the tuned cells were ordered topographically along the rostrocaudal axis of the BIN, although this representation is considerably more scattered than that in the SC. In contrast to the SC, we observed no systematic variation in the distribution of near-threshold best azimuths, which were instead concentrated around the interaural axis in the contralateral hemifield. The azimuth tuning of individual units in the BIN was generally broader at both sound levels than that in the SC. Many units also were tuned for the elevation of the sound source (48% for supra-, 77% for near-threshold stimulation), but there was no evidence for topographic order in the distribution of preferred elevations within theBIN. These results suggest that the BIN sends inputs to the SC that are already selective for sound azimuth and elevation and that show some degree of topographic order for sound azimuth. These inputs then presumably are sharpened and their topography refined by a mechanism that is likely to involve convergence of other inputs and activity-dependent fine tuning of terminal connections, to result in a precise two-dimensional map of auditory space in the SC.

INTRODUCTION

Auditory neurons in the deeper layers of the mammalian superior colliculus (SC) have spatially tuned receptive fields that are arranged systematically to form a two-dimensional map of auditory space across the horizontal extent of this midbrain nucleus (King and Hutchings 1987; Middlebrooks and Knudsen 1984; Palmer and King 1982). Studies in anesthetized animals have shown that the map of auditory space is aligned topographically with maps of the visual field and of the body surface (Stein and Meredith 1993). The registration of different modality receptive fields also appears to be maintained at least partially in awake animals capable of making independent movements of their eyes and pinnae (Hartline et al. 1995; Jay and Sparks 1987; Peck et al. 1993). This arrangement, in which different sensory representations share the same coordinates in the SC, is thought to provide an efficient means of enabling neurons to integrate spatial cues they receive from different sensory channels and of allowing a novel target, irrespective of its modality, to elicit an orientation response through the same premotor pathways (Knudsen and Brainard 1995; Stein and Meredith 1993).

Although the pathways conveying visuotopic and somatotopic representations to the mammalian SC have been well documented, the neural circuitry underlying the synthesis of the auditory space map is understood very poorly. In contrast to the other modalities, a topographic representation of auditory space has to be computed centrally by neurons that are differentially sensitive to the auditory localization cues that result from the interaction of sound with the head and ears (reviewed by King and Carlile 1995). The barn owl SC or optic tectum receives a point-to-point projection from a nucleus that is thought to be homologous to the external nucleus of the inferior colliculus (ICX) (Knudsen and Koni- shi 1978), and both midbrain structures contain similar maps of auditory space (Knudsen 1982; Knudsen and Konishi 1978). The processing steps that take place in the avian brain stem leading to the formation of a place code for sound location in the ICX also have been described in some detail (Konishi 1995).

We recently have obtained neuroanatomic data showing that, in ferrets, the brachium of the inferior colliculus (BIC), and particularly its nuclear region (BIN), provides the largest projection from a putative auditory structure to the deeper layers of the SC (Jiang et al. 1993; King et al. 1992; King et al. 1997a). Projections from the BIN to the SC also have been reported in mice (Wallace and Freder, 1989), hamsters (van Buskirk 1983), and cats (Edwards et al. 1979; Kudo et al. 1984). At its rostral end, the BIN merges with the principal part of the medial geniculate nucleus (MG) and courses caudally to lie lateral to and parallel with the deeper layers of the SC. The caudal end of the BIN terminates at approximately the same level as the rostral pole of the ICX, whereas the more lateral fibrous region of the BIC extends further back to overlap with the major part of the ICX (see Fig. 1). The area that we have designated the BIN (see Berman 1968; King et al. 1997a,b) appears to correspond to the accessory region described by Morest and Oliver (1984) and consists of neurons with a tectal morphology. Most of the BIC neurons that were labeled retrogradely after injections of either horseradish peroxidase or of fluorescent microspheres into the ferret SC were found in the accessory part of the nucleus (King et al. 1997a). A
Here we describe the results of a mapping study of the ferret BIN in which we examine the responses to free-field stimuli of units recorded in different regions of this nucleus. Our results show that, like in the SC, a large proportion of units recorded in the BIN are tuned both in azimuth and elevation to single regions of space and that, at least for broadband stimuli presented at relatively high sound levels, the preferred sound azimuths of these units vary systematically along the rostrocaudal axis of the BIN.

Preliminary accounts of some of these results have been published in abstract form (King and Schnupp 1995; Schnupp and King 1995).

**METHODS**

In this study, recordings were made from five pigmented adult ferrets. For full details of the surgical preparation, see King and Hutchings (1987) and King et al. (1994). Briefly, the animals were anesthetized with an initial intramuscular injection of 0.9% wt/vol alphaxalone, 0.3% alphadolone acetate (Saffan, Glaxovet) at a dose of 2 ml/kg. During surgery, additional doses of Saffan were administered intravenously as required. Body temperature was monitored rectally and held constant at 39°C. A craniotomy was performed to expose the cortex overlying the right BIN and SC, and the skull fixed to a minimal head holder that was attached to a supporting post behind the animal. Care was taken to restore the natural position of the pinnae after craniotomy. The left eyelids were removed, and the eye protected with a zero refractive power contact lens to allow the mapping of visual receptive fields in the SC, which were used as landmarks to guide electrode penetrations into the BIN through the intact cortex. To prevent eye movements, the animals were paralyzed with 12 mg intravenous gallamine triethiodide (Flaxedil) and artificially ventilated with room air supplemented with 95%O2-5%CO2. During recording, paralysis and anesthesia were maintained by a continuous infusion of 20 mg·kg⁻¹·h⁻¹ Flaxedil and 1 mg·kg⁻¹·h⁻¹ pentobarbital sodium (Nembutal) in Locke’s solution. The heart rate, electrocardiogram, electroencephalogram, and end-tidal CO2 were monitored continuously as a means of assessing that the animals were maintained in an adequate and stable level of anesthesia.

Extracellular recordings were made in an anechoic chamber. Visual stimuli consisted of 100-ms light flashes delivered from a 1-cm-diam LED. Digitally generated broadband noise bursts (30–30,000 Hz, 100-ms duration with a 5-ms rise/fall time) were delivered from a hoop-mounted speaker (Kef T27), which allowed stimuli to be presented from almost any direction relative to the animal’s head. The noise was regenerated before every presentation, i.e., we did not use “frozen” noise stimuli. Bursts were presented with an interstimulus interval of ≈1.5 s. The output of the free-field speaker was calibrated to dBS sound pressure level (SPL) with a microphone placed in the anechoic chamber at the position occupied by the center of the animal’s head during the electrophysiological recordings.

Single-unit recordings were made using conventional tungsten-in-glass microelectrodes. The electrode was advanced stereotaxically into the brain stem through the intact overlying occipital cortex. Penetrations were made at a slight angle (~10° from the vertical to the lateral side) to allow the electrode to run roughly parallel to the maximal dorsoventral extent of the BIN. The recorded signal was band-pass filtered (500–5,000 Hz), amplified and digitized at a rate of 25 kHz. We isolated single units from the digitized signal using our own spike-sorting software. This software is a Windows-based program that implements the now fairly widely used “cluster cutting” paradigm (see e.g., Gray et al. 1995). Spike features (e.g., spike amplitude vs. spike duration or the amplitude of the first vs. that of the second phase of the
spike) are measured on-line and plotted on a Cartesian coordinate system. Action potentials from a single unit tend to form clusters in these plots, and clusters can be delineated by the experimenter. Figure 2 shows one of these “feature space” scatter plots. Three clusters can be distinguished. The action potential shapes from each of the clusters are shown superimposed in the plots on the right.

We first made vertical electrode penetrations into the SC and used the location of the visual receptive fields of multiunit activity recorded in the superficial layers as a means of finding the BIN, which lies lateral and ventral to this region of the SC. Once the lateral edge of the SC had been found, as indicated by the elevation coordinates of the visual receptive fields, subsequent electrode tracks were made 500–1,000 µm further lateral. Figure 1 illustrates the location of the BIN relative to the SC and other brain stem nuclei in a series of camera lucida drawings. The borders of this nucleus were identified on the basis of anatomic studies of the IC in the cat (Berman 1968; Morest and Oliver 1984) and especially from the distribution of retrogradely labelled neurons found after injections of horseradish peroxidase or fluorescent microspheres in the SC (King et al. 1997a). For each auditory unit, we first estimated its threshold with the loudspeaker positioned either near the interaural axis contralateral to the recording site or at a position that had yielded strong responses for other units in the same track. We then determined the unit’s azimuth response profile at two sound levels, typically 5–15 and 25–30 dB above unit threshold, by moving the speaker in 20° steps around the animal’s horizon from positions 160° contralateral to 160° ipsilateral to the recording site. The discharge of the unit was measured in response to 20 stimuli presentations at each speaker position. After the first azimuth run, we typically redetermined the unit thresholds at the speaker azimuth that had given the strongest response. We also determined the unit’s elevation response profile by moving the speaker from 60° below to 80° above the audio-visual horizon at the azimuthal position that gave the strongest response.

Recording sites were marked with small electrolytic lesions (5 µA for 5 s). At the end of the recording session, typically after ~40 h of recording, the animal was overdosed with pentobarbital sodium (Euthatal, intravenous) and perfused through the heart with phosphate-buffered saline followed by 10% formal saline. The brain stem was removed, cryoprotected with 30% sucrose, cut in 50-µm coronal sections, and Nissl-stained for the histological reconstruction of the recording sites. Figure 3 shows one of these Nissl-stained sections with electrolytic lesions in the SC and the BIN.

By varying the pattern of lesions (and by making some in the adjoining SC as additional landmarks), we were able to distinguish all the electrode penetrations reconstructed from the coronal sections. The location of the lesions (and, if visible, the electrode tracks themselves) were used together with the microdrive readings to identify both the section containing a given recording site, and, within that section, the distance of the recording site from the dorsal and lateral borders of the BIN. Occasionally, neighboring lesions merged, making it more difficult to identify the dorsoventral coordinates of the recording sites. Units recorded at those sites were therefore not included when considering the variation in preferred sound directions along this dimension of the nucleus.

RESULTS

We recorded from a total of 176 units at 61 recording sites in 35 electrode penetrations. Of these, 112 units from 22 penetrations were adjudged to be located in the BIN and 6 units from two penetrations were found to be in the lateral, interstitial region of the BIC. The remaining units were located in deep SC, MG, or ICX. Allocation of units to the BIC was made on the basis of histological reconstructions, using criteria established in the anatomic study by King et al. 1997a. The dorsal, ventral, and lateral borders of the BIN are easy to identify in Nissl-stained sections. However, this is not the case for the medial border with the SC or the ICX, so this was determined on the basis of the location of neurons retrogradely labeled by large injections of either horseradish peroxidase or of fluorescent microspheres into the deeper layers of the SC (see King et al. 1997a). Units recorded in electrode penetrations that ran very close to this border could not be unambiguously localized to the BIN and were therefore not included in our sample.

Temporal response profiles

All single units recorded in the BIN showed some level of spontaneous activity and most had relatively complex temporal response profiles. Because the responses were often quite labile, we generated poststimulus time histograms (PSTHs) by pooling all the data for each unit. We broadly subdivided the PSTH patterns that we observed into three classes: about 42% of the units showed onset responses, which consisted of a single evoked spike or burst of spikes occurring typically between 8 and 30 ms after arrival of the stimulus at the ears. A further 51% gave multipeaked responses, in which the PSTH additionally showed a second and often a third peak separated by periods in which the activity fell back to spontaneous levels. The time course during which the later peaks appeared varied markedly
of spikes in an equivalent length of time in the spontaneous window. The response strengths of individual neurons ranged, at the optimal stimulus position with stimuli ~25 dB above threshold, from 0.5 to 13 evoked spikes per presentation. Strongly and weakly responsive cells tended to occur in clusters. We did not observe any systematic order in the location of these clusters within the BIN.

The distribution of unit thresholds is shown in Fig. 5. The mean threshold was ~30 dB SPL, the mode ~35 dB SPL, and 75% of the units had thresholds between 17 and 44 dB SPL.

Response latencies

For this study, we considered it very important to arrive at a good estimate of the response latencies for BIN units. As we were interested in the possibility that these neurons provide a major source of input to the auditory SC, we would predict that the latencies in the BIN are on average slightly shorter than those in the SC. Furthermore, it is well established that the temporal characteristics of a response in the auditory system may vary systematically with stimulus level or position, and such changes may contribute to the neural code. These changes may range from simple latency shifts, as documented, for example, in the barn owl optic tectum (Knudsen 1984) to complex temporal codes like those described by Middlebrooks et al. (1994) in the anterior eustachian sulcus (AES) of the cat.

The variation in temporal firing pattern with stimulus position is shown for two units in Figs. 6 and 7. The raster plots in Fig. 6 are from the unit whose PSTH is shown in Fig. 3. Nissl-stained coronal section through the brain stem showing 3 electrolytic lesions (*). Lesions mark 2 penetrations, 1 in the superficial SC, 1, marked with a double lesion, in the BIN. This section is taken from a position about midway through the rostrocaudal extent of the BIN. Scalebar (bottom) represents 1 mm. SCS, SC superficial layers; SCD, SC deeper layers; PAG, periaqueductal gray.

among these units, with the last peak extending 70–300 ms beyond stimulus onset. Sustained responses (characterized by a more or less continuous period of increased firing lasting typically from 8 to 130 ms, and occasionally up to 200 ms, after stimulus onset) were rarer, at ~7%. Figure 4 gives representative examples of these response types.

Response strengths and thresholds

The often high spontaneous activity and the complex and variable temporal response patterns complicate the precise determination of the response strengths, latencies, or thresholds for these units. To calculate the firing rate, we set a “response window” for each unit to cover all the response peaks in the pooled PSTH for that unit and set a “spontaneous window” arbitrarily to cover the period from 500 to 1,000 ms after stimulus onset. The number of evoked spikes then was calculated from the mean number of spikes in the response window per presentation minus the mean number

FIG. 3. Nissl-stained coronal section through the brain stem showing 3 electrolytic lesions (*). Lesions mark 2 penetrations, 1 in the superficial SC, 1, marked with a double lesion, in the BIN. This section is taken from a position about midway through the rostrocaudal extent of the BIN. Scalebar (bottom) represents 1 mm. SCS, SC superficial layers; SCD, SC deeper layers; PAG, periaqueductal gray.


These histograms were generated from the pooled data recorded over all stimulus positions and plotted with a binwidth of 5 ms. Timing and the duration of the stimulus (100-ms noise burst) are indicated by the solid bars below the x-axes.
FIG. 5. Distribution of unit thresholds in the BIN.

4B, which had three peaks in its PSTH profile. This response includes a very broadly tuned, longer latency component that begins $\sim 20$ ms after stimulus offset. The latency of this third peak in the temporal response profile is consistent with this being an offset response. However, because we did not vary the stimulus duration, we are unable to confirm this possibility. Figure 7 is from a two-peaked unit, with a primary, sharp onset response centered on $\sim 12$ ms, and a broader, second component centered on 80 ms.

For both units, there may be small variations in the latency of the onset of the first response peak with azimuth. For example, the weaker responses produced in the ipsilateral hemifield by the unit shown in Fig. 7 appear to have a longer latency than those evoked by more effective contralateral stimulation. Other aspects of the temporal firing patterns also varied systematically with sound azimuth. For the unit illustrated in Fig. 7, the second peak of the PSTH, from $\sim 50$ to 110 ms, was present only for stimulus azimuths between $-40^\circ$ and $-120^\circ$ azimuth. These stimulus-dependent variations resemble the firing patterns described by Middlebrooks et al. (1994) in the AES of the cat and may form the basis of a temporal code for sound location.

On the basis of the number of evoked spikes, we classified the unit in Fig. 6 as tuned to $100^\circ$ azimuth contralateral to the recording site ($-100^\circ$), and the unit in Fig. 7 as tuned to $-40^\circ$. The spatial tuning for the various response components of these units is illustrated in Fig. 8. In this figure, the average spike counts (after subtraction of the spontaneous rate) for each of the temporal components of the responses are plotted against azimuth using a polar coordinate system. The different components of the three-peaked response illustrated in Fig. 6 appear to vary somewhat in their spatial tuning (Fig. 8, top). On the other hand, very similar spatial
tuning was exhibited by the first and second components of the response of the unit from Fig. 7 (Fig. 8, bottom).

The distribution of first spike latencies at any given loudspeaker position frequently was spread and skewed by the combination of weak responses and high spontaneous activity. We therefore based our estimates of onset latency on the timing of the first peak in the PSTH generated from the pooled data for an entire azimuth run carried out at a single sound level. These suprathreshold response latency values are summarized in Fig. 9 in histogram form. The distribution of latencies shown there is extremely similar to that reported for the SC at 20 dB above threshold in Fig. 3C of King and Hutchings (1987).

**Azimuth response profiles**

We recorded suprathreshold azimuth response profiles from 83 units in BIN as well as from 1 unit in the more lateral interstitial region. Response profiles also were obtained from nine units on the border between SC and BIN and from two units in the ICX. The mean sound level (±SD) at this suprathreshold condition for the cells in the BIN was 28 ± 4 dB with respect to unit threshold. In the near-threshold condition, we recorded azimuth profiles of 64 units in the BIN and 4 units in the interstitial region at a mean sound level of 6 ± 4 dB, as well as from 3 units on the SC/BIN border, 5 units in deep SC, and 6 cells in auditory thalamus. In the following, we present data from the units found in the BIN and in the interstitial part of the BIC only.

To classify these auditory units according to their spatial tuning characteristics, we assigned each azimuth response profile to one of five categories. Tuned units show only a single peak in their response profile, whereas bilobed cells had two peaks and complex cells more than two. In each case, a peak is defined as a region where the response reached 80% of maximum, flanked by regions of 40% or more on either side and with the flanking regions not more than 160° apart. Broad or hemifield units have a peak which, at 40% of the maximal response, spans >160°, whereas omnidirectional units respond with 40% of the maximum response regardless of speaker position. Although somewhat arbitrary, these same criteria for assigning peaks and valleys were used in several previous studies of the ferret SC (e.g., Schnupp et al. 1995b) and were adopted here to facilitate a direct comparison with our SC normal data set. The response profiles were sorted by computer to avoid observer bias and examples of BIN units in each category are given in Fig. 10A. Our definition of a peak in the response profile allows a “tuned” unit to respond relatively well over a fairly large azimuthal range. However, the great majority of these units had peaked response profiles, with the maximum response occurring over a narrow region of space and usually at a single loudspeaker position. Further examples of azimuth profiles, which illustrate this point, are shown in Fig. 11.

Figure 10B shows the relative proportions of each of the response profile types at near- and suprathreshold sound levels in the BIN for the whole study population. We found a marked increase in the proportion of broad and a decrease in the proportion of tuned cells at higher sound levels. A χ²-test on these observed proportions indicates that these differences are statistically highly significant (P < 0.01). In comparison, in the SC of the same species tested under suprathreshold conditions 77% of units have been reported to be tuned, 6% bilobed, and 17% broad (Schnupp et al. 1995b).

Figure 11 shows the suprathreshold response profiles obtained at six recording sites in the BIN of a single animal.
The response profile plots are arranged in tabular form, one row per recording site, with different response profile types arranged in columns. The units classified as tuned typically responded to noise stimuli throughout much of the contralateral hemifield, but each nevertheless had a well-defined best position where the maximum response was obtained. These best positions showed some tendency to shift laterally at progressively more caudal recording sites. In contrast most of the ‘‘broad’’ units, which were not ascribed best positions, responded almost equally well over an entire hemifield. However, some of these units did exhibit a broad spatial preference that matched that of tuned units recorded at the same location within the BIN. For example, unit u006, recorded in the most rostral penetration, responded most strongly to a region just contralateral of the anterior midline, whereas units u031 and u054 were maximally responsive over more lateral regions of this hemifield. Bilobed or complex cells appeared to have several noncontiguous best positions.

The increase in the proportion of broad cells at the expense of tuned cells noted in Fig. 10B is probably a manifestation of a general trend of auditory receptive fields in the BIN to expand with increasing sound level, a trend that also has been documented in the SC. Studies in several species, including our own previous work on the SC, have assessed the sharpness of spatial tuning by measuring the 50% bandwidth of the response profile. The expansion of the receptive fields with increasing sound level is illustrated in Fig. 12, which shows histograms of the 50% bandwidths of the azimuth profiles of all the units recorded in the BIN and compares them with those reported for the SC. Note that, like King and Hutchings (1987), we include all regions of the response profile exceeding the 50% level in the measurement of the bandwidth, whether or not these regions are classifiable as peaks according to the criteria discussed above. Figure 12, A and B, shows the bandwidths observed in the BIN at supra- and near-threshold sound levels, respectively, whereas Fig. 12, C and D, shows the corresponding data for the SC (from King and Hutchings 1987). In both nuclei, we observed broader tuning at higher sound levels. SC units tend to be tuned more sharply for azimuth than those recorded in the BIN. Mann-Whitney U tests were used to confirm that these differences are statistically significant ($P < 0.01$ for all pairwise comparisons of data from each nucleus at different sound levels or between nuclei at the same sound levels).

BIN units thus resemble SC units in the types of azimuth profiles observed and in the fact that their azimuth profiles tend to expand with increasing sound level. But, apart from being typically more broadly tuned, BIN units differ markedly from SC units in another important respect: although the auditory best positions of units in the SC generally do not vary with sound level, those in the BIN frequently do. When we compared the near- and suprathreshold best positions for 32 units in the BIN that were classified as tuned at both sound levels, we found that the differences in the best positions were distributed approximately normally, with a mean near zero, but with a standard deviation as high as 66.5°. These differences are shown in Fig. 13 in histogram form.

Topography of azimuth tuning

To examine whether the preferred sound directions vary systematically across the BIN, we first constructed a map of all our recording sites within the nucleus from measurements taken from the series of coronal sections through the brain stem that were prepared after recording. This map is shown in Fig. 14. For the purpose of generating this map, we treated the BIN as an essentially two-dimensional structure. Its longest dimensions are the rostrocaudal and dorsoventral axes, the dorsoventral extent increasing from rostral to caudal (see Fig. 1). Medially, it is widest near the rostral end, measuring 1–1.5 mm across, whereas most of the caudal third comprises a thin sheet of interstitial BIC, which lies lateral to the ICX. It is exceedingly difficult to make re-
cordings in this interstitial region because the electrode is very likely to enter the ICX by mistake. Moreover, very few neurons are labeled there after tracer injections in the SC (see King et al. 1997a). The dorsoventral measurements that were used in the construction of this map were taken along the axis of the maximal extent of the nucleus on each coronal section. The angle of this axis relative to the vertical varies from ~5 to 20° from the caudal to the rostral end of the BIN.

In an attempt to visualize the representation of sound azimuth in the BIN, we averaged the suprathreshold best positions for all the tuned units recorded at each site and then varied the darkness of the symbol corresponding to each recording site to obtain a map of sound azimuth for the BIN. This is shown in Fig. 15. Although there is no strict progression, there does appear to be a trend for more posterior azimuths to be represented more caudally. To test whether this trend is statistically significant, we performed linear regression analyses of best azimuths versus unit position measured along axes running at various angles through this map. We found that an axis running at an angle of +18° relative to the horizontal gave the highest $R^2$ value of 0.34. This axis can be thought of as an “azimuth axis” and is shown in Fig. 15 as a stippled line.

Figure 16A shows scatter plots of best azimuths versus recording site measured along the 18° optimal azimuth axis. Linear regression lines and $R^2$ values for these data also are shown. The spread of the data points around this azimuth axis is quite high, indicating that the azimuth topography in the BIN is either fairly scattered or poorly described with this type of linear analysis. Nevertheless, the regression is highly statistically significant ($n = 33, P = 0.0003$). In Fig. 16B, we show corresponding data for the SC that were collected during a number of previous studies. King and Carlile (1993) found that the azimuth topography in the SC is best described by a second order polynomial rather than by a linear regression. This polynomial fit for the SC azimuth representation, shown in Fig. 16B, is preferable because the
The topography of the auditory map appears to be largely shaped by that of the visual map in the SC superficial layers. This visual map, like many other visual maps in the brain, devotes a disproportionately large area of tissue to the representation of central vision. Accordingly, a relatively large proportion of auditory SC units are tuned to positions on and near the anterior median plane. In the BIN, in contrast, units tuned to 0° azimuth were not encountered.

The bilobed response profiles were allocated two best positions. For nearly every BIN unit, both best positions were contralateral to the recording site. These are plotted against the rostrocaudal location of the recording electrode in Fig. 17. Because bilobed responses may exhibit appropriate, albeit ambiguous, tuning (see Carlile and King 1994), we looked for systematic order in their distribution by performing a linear regression analysis based on the best azimuth of the peak that was closest to the regression line fitted to the best positions for the tuned units. The slopes of the two regression lines were very similar, but the value of $R^2$ for the “appropriate peaks” of the bilobed response profiles failed to reach significance ($P < 0.07$).

Although we found some evidence for a topographic supra-threshold map of sound azimuth, we found a rather different picture when we applied the same analysis to our near-threshold azimuth dataset. Figure 18 shows a map of mean best positions for the near-threshold azimuth dataset and is analogous to Fig. 15 for suprathreshold data. Two of the 44 tuned cells from 25 recording positions showed near-threshold tuning to positions in the posterior part of the ipsilateral side of space (indicated by *), whereas the tuning under suprathreshold stimulation was exclusively contralateral. The occurrence of these ipsilateral best positions is interesting but awkward from the point of view of the linear regression analysis described above, not just because regression analysis weights outliers very heavily, but because we would have to use circular, rather than linear, statistics if the data are not restricted to a well-delimited portion of space. For these reasons, we decided to exclude these two data points from the linear regression analysis. The remaining near-threshold data revealed no significant correlation between best azimuth and anatomic position along any axis through the BIN. In other words, the BIN displays a topographic map of sound azimuth only under suprathreshold stimulus conditions.

Figure 19A is equivalent to Fig. 16A and plots best azimuth positions of tuned cells against unit position along the anatomic axis shown in Fig. 15. There is no significant correlation. Instead best positions appear loosely clustered in the anterior quadrant of the contralateral hemifield, particularly just in front of the interaural axis. In this respect, the BIN behaves very differently from the SC (see Fig. 19B), where best azimuths remain topographically arranged under near-threshold stimulation, and the topography of the representation is statistically indistinguishable from that at suprathreshold sound levels (Carlile and King 1994).

**Elevation tuning**

We also recorded elevation receptive field profiles in the BIN by recording responses at a series of speaker positions varying in elevation from 60° below to 80° above the animal, while keeping the azimuth of the speaker constant at or near the unit’s best azimuth. The elevation profiles then were classified as tuned, bilobed, or broad according to criteria similar to those used to characterize the azimuth response profiles. The scheme used for classifying elevation profiles differs slightly from that used for azimuth profiles to account for the fact that, although an azimuth profile describes a full
FIG. 15. Map of mean best positions for each recording site at which suprathreshold azimuth data were obtained. Recording sites are plotted in the same way as in Fig. 14, but different symbols are used to show the average azimuth preference observed at that site as a means of deriving the overall azimuth preference at each position. Recording sites with a preference for frontal azimuths were found mainly in the rostral part of the nucleus, whereas sites with a preference for posterior stimulus positions were more common in the caudal region. Regression analyses showed that this tendency was significant (see text).

FIG. 14. Map of recording sites in the BIN. Dorsal and the ventral borders of the BIN. They were derived by fitting polynomials to serial reconstruction measurements from 3 animals. ●, position of the recording sites within the nucleus. x-axis gives the rostrocaudal position of each recording site. Measurements along the y-axis (dorsoventral) were taken along the maximal dorsoventral extent of the nucleus and normalized to compensate for interanimal size variations.

circle, an elevation profile only describes a semicircle (or meridian) in the hemifield contralateral to the recording site. A peak (i.e., a region of $\approx 80\%$ of the maximal response) in the response profile therefore could be delimited either by a position with $\approx 40\%$ of the maximal response or by one of the poles of the coordinate system. For broadly tuned units, the response does not fall to $\approx 40\%$ of the maximum value at any position. A tuned elevation profile shows at least one such valley but no second peak, whereas bilobed units gave relatively high responses ($\approx 80\%$ of maximum) on either side of a low response position. Figure 20 shows representative examples of these types of elevation profiles recorded in the BIN.

We classified suprathreshold elevation profiles recorded from 42 units and near-threshold elevation profiles from 30 units. Of the suprathreshold profiles, 20 (48\%) were tuned, 4 (10\%) were bilobed, and the remaining 42\% were broad. In the near-threshold condition, 23 profiles (77\%) were tuned, 1 (4\%) was bilobed, and the rest (20\%) were broad. As with the azimuth profiles, we found that the proportion of tuned cells was higher for the near-threshold condition.

Because a large proportion of units in the BIN appeared to be elevation sensitive, we looked for evidence for a topographic representation of sound source elevation. At suprathreshold stimulation, the best elevations ranged from $+60^\circ$ to $-60^\circ$, although the majority (14/20, 70\%) were tuned to positions between $40^\circ$ above and $20^\circ$ below the audiovisual horizon. The range of best elevations observed with near-threshold stimulation was similar, with a total range from $-60^\circ$ to $+40^\circ$ and 69\% (16/23) of the units tuned to elevations between $-40^\circ$ and $+20^\circ$. Figure 21 shows the distribution of mean best elevations (averaged over all units recorded at each of the sites shown) over the rostrocaudal and the dorsoventral extent of the BIN, for both suprathreshold and near-threshold stimulus levels. In the SC, sound elevation is mapped mediolaterally, a dimension not shown in this figure. However, our recent anatomic study failed to reveal any segregation across the mediolateral axis of the BIN in the distribution of retrogradely labelled projection neurons after injections of different tracers into the medial and lateral halves of the SC (King et al. 1997a,b). We therefore have illustrated the location of the recording sites with respect to the same sagittal view of the BIN used in
FIG. 16. Azimuth topography plots. A: suprathreshold best azimuth for all the tuned units is plotted against position within the nucleus, measured along the stippled axis shown in Fig. 15. B: suprathreshold best azimuth topography plot for the SC. Correlation between azimuth tuning and anatomic position in the BIN is weak but significant, accounting for ~34% of the variance observed. It appears that the azimuth topography in the BIN is considerably more scattered than that in the SC.

Figs. 15 and 17 rather than its much smaller mediolateral axis. We failed to find a significant variation in best elevation along any axis within the BIN at either sound level, irrespective of whether we considered the stimulus positions evoking the maximum response for all units or restricted this analysis to the best positions of the tuned units only.

For each unit, we first obtained an azimuth profile at a fixed elevation and then measured the elevation tuning at the best azimuth (where available) for that unit. The same approach was adopted previously for the SC, revealing the presence of topographic maps of both sound azimuth and elevation (King and Hutchings 1987). However, if the receptive fields of BIN units are irregular in shape or oriented

FIG. 17. Best azimuths of bilobed units plotted against recording site within the BIN, measured along the azimuth axis shown in Fig. 15. Two best positions for each unit are shown by a filled circle and horizontal bar joined by a vertical line. ●, best azimuth that lies closer to the regression line fitted to the best positions of the tuned units; ——, regression line for these “appropriately tuned” lobes vs. unit position, and its associated $R^2$ is provided (top right). ———, regression line obtained for tuned units (Fig. 16A). An equivalent plot is not provided for the SC as only 3 units in that nucleus were classified as having bilobed azimuth response profiles.

FIG. 18. Map of mean best positions within the BIN for the near-threshold azimuth dataset. Layout of this figure is analogous to that of Fig. 15. In contrast to the suprathreshold data, the near-threshold responses shown here reveal no systematic variation of preferred sound azimuth within the BIN.

FIG. 19. Near-threshold azimuth topography plots. These graphs are analogous to those in Fig. 16. A: near-threshold best positions do not vary systematically with recording site position but cluster just in front of the interaural axis on the contralateral side. B: in contrast, the near-threshold best azimuths of SC units are ordered topographically.
FIG. 20. Examples of the 3 classes of elevation profiles observed.

away from the vertical axis, it is possible that we would have obtained a different estimate of best position by constructing spatial response profiles away from the best angle in the other dimension.

Responses to visual stimuli

Previous studies of auditory cells in the deeper layers of the SC have shown that a large proportion of units there are also responsive to visual stimulation. Because there have been several reports of projections from the SC to the IC (Adams 1980; Sun and Jen 1989), we examined, for a small subset of our sample, whether units in the BIN also can be driven by visual stimuli. Fifteen auditory units from seven different recording sites in the BIN were tested with a flashing LED placed in front of the eye contralateral to the recording site. Three (20%) of these units were bimodal and also showed weak responses to light. At one recording site, we also observed a unimodal visual unit. As far as we could tell from the histological reconstructions of our electrode penetrations, all of these units definitely were located in the BIN rather than the deep layers of the SC. This sample is limited both in the number of units tested and in that only a stationary flashing light stimulus was used, but it nevertheless shows that visual responses do occur in the BIN, although they appear to be rarer and weaker than those in the SC.

DISCUSSION

The aim of this study was to examine whether the responses of auditory units in the BIN of the ferret represent sound location in a systematic manner. We found that the majority of units exhibited a clear preference for particular regions of space. Their preferred sound directions covered a wide range of azimuths and elevations within the contralateral hemifield. At sound levels of ~25 dB above unit threshold, there was a tendency for the best azimuths to vary topographically, with anterior positions represented predominantly in rostral BIN and more peripheral positions represented in caudal regions of the nucleus. These findings are consistent with a recent anatomic study of the auditory inputs.
to the ferret SC (Jiang et al. 1993; King et al. 1992, 1997a,b). Together, these studies indicate that the BIN is a major source of auditory input to the ipsilateral SC and suggest that this nucleus may play an important role in providing information about the azimuthal location of sound sources.

If the BIN does project systematically to the SC, we would expect to find that these units have slightly shorter latencies than those in the SC. An accurate assessment of response latency is problematic because of the nonstationary spontaneous activity and the weak and labile responses often associated with auditory units recorded in both structures. Based on the summed PSTHs for each unit, we found that the range of onset latencies of BIN units is very similar to and, on average, slightly shorter than that previously reported for the ferret SC (King and Hutchings 1987). This is what we would expect if the BIN does project systematically to the SC. Nevertheless, in both cases, a wide range of onset latencies was observed. Moreover, it seems unlikely that all the neurons in the BIN project to the SC. Simultaneous recordings from neurons in both nuclei therefore will be required to describe further the functional role of connections between the SC and the BIN.

Very little is known about the physiological properties of neurons in the BIN. This nucleus is innervated by the central nucleus of the IC (ICC) and the ICX (Kudo and Niimi 1980) and has been reported to project to the MG (Kudo et al. 1984) and primary auditory cortex (Winer 1984) as well as the SC (Edwards et al. 1979; Jiang et al. 1993; King et al. 1992, 1997a,b; Kudo et al. 1984; van Buskirk 1983; Wallace and Fredens 1989). Several groups have examined the responses of units recorded in the ICC and ICX to sounds presented in the free field. However, a direct comparison with our results is difficult because most of these studies employed best-frequency tone stimuli rather than noise bursts. Nevertheless, units that are azimuth selective with noise have been reported in both the ICC (Aitkin and Martin 1987; Aitkin et al. 1984) and ICX (Aitkin et al. 1984; Binns et al. 1992). In high-frequency regions of the cat ICC, Aitkin and Martin (1987) found that the best azimuths clustered within a 20° region contralateral to the anterior median plane. On the other hand, a much greater range of best azimuths, covering almost the entire contralateral hemifield, has been reported in the guinea pig ICX (Binns et al. 1992). In the only other free-field investigation to be carried out on the BIN, Aitkin and Jones (1992) noted that this nucleus in the cat contains a greater proportion of spatially tuned units than either the ICC or the ventral nucleus of the MGN. We found that both the BIN and the SC contain large proportions of units that appear tuned to specific sound locations, suggesting that these midbrain nuclei, together with the ICX, may be particularly concerned with representing auditory space.

The range of best azimuths and elevations exhibited by BIN units closely resembles that previously described for the ferret SC (Carlile and King 1994; King and Hutchings 1987; King et al. 1994). However, there are a number of differences between the nuclei in the topographic organization of these preferred sound directions. In the SC, sound azimuth is mapped along the rostrocaudal axis of the nucleus, whereas the best elevations of the units vary systematically across its mediolateral axis. Moreover, this place code for auditory space appears to be level-independent as changing the sound level has a negligible effect on the best positions of the auditory units (Carlile and King 1994; King and Carlile 1994; King and Hutchings 1987). In contrast, increasing the sound level often altered the spatial response profiles of BIN units, and only one-third of the tuned units in the BIN exhibited shifts in best azimuth of ±20° with increasing sound level. The great majority of the best azimuths measured at near-threshold sound levels were clustered around or just in front of the interaural axis in the contralateral hemifield. This is quite unlike the precise topographic order of best azimuths recorded at corresponding sound levels in the SC (see Fig. 19). However, at sound levels of ~25 dB above threshold, we did observe a systematic change in best azimuth along the rostrocaudal axis of the BIN, although this representation certainly was scattered more than that found in the SC. Despite the presence of some topographic order in the representation of sound azimuth, we failed to find any indication for a systematic variation in elevation tuning within the BIN. In fact, these findings are entirely consistent with the results of the recent anatomic study by King et al. (1997a), which provided no evidence for distinct patterns of input from the BIN to the medial and lateral halves of the SC, where superior and inferior sound locations are represented, respectively. On the other hand, the rostral and caudal regions of the SC are innervated primarily by corresponding regions of the BIN, and both nuclei show at least some topographic organization in the representation of sound azimuth along this axis.

Some auditory neurons in the SC exhibit sharp frequency tuning, whereas others have broad, multipeaked tuning curves, with a preference for high frequencies (Carlile and Pettigrew 1987; Hirsch et al. 1985; King and Palmer 1983; Middlebrooks 1987; Wise and Irvine 1983) and derive their azimuthal spatial selectivity from a combination of monaural spectral cues and interaural level differences (Carlile and King 1994; Hirsch et al. 1985; King et al. 1994; Middlebrooks 1987; Palmer and King 1985; Wise and Irvine 1985). Monaural spectral cues provided by the outer ear vary with both the horizontal and vertical location of the sound source (Carlile and King 1994) and appear to be largely responsible for the topography observed in the SC at near-threshold sound levels, which is unaltered by occlusion of or cochlear ablation in the ipsilateral ear (King et al. 1994; Palmer and King 1985). In contrast to the SC, the lack of topographic order in the near-threshold representation of sound azimuth in the BIN suggests that neurons there may not be differentially sensitive to direction-dependent features present in the spectral transfer function of the auditory periphery. Although we did not use best-frequency tones, it does appear that the near-threshold best azimuths of BIN units cluster in the region of the acoustical axis of the contralateral pinna (King and Carlile 1994), suggesting that their spatial selectivity, as in the ICC (Moore et al. 1984; Semple et al. 1983), may be attributed to the region of maximum gain provided by the outer ear.

The thresholds of auditory units in the BIN and the SC are very similar (compare Fig. 5 with Fig. 1 of King et al. 1994), indicating that the same localization cues are likely to be available at corresponding sound levels. Aitkin and
Jones (1992) reported that units in the BIN of the cat prefer high-frequency sounds, suggesting that, like in the SC, interaural level differences are the principal binaural cue underlying the topographic order that we have observed at higher sound levels. Although the binaural response properties of units in the BIN have yet to be described, it is of interest to note the absence in our data of units tuned to the anterior midline. Many units in rostral SC are tuned to this location, probably on the basis of facilitatory interactions between the inputs from the two ears (Middlebrooks 1987; Wise and Irvine 1985). Although the paucity of units tuned to 0° azimuth may reflect the difficulty of recording from the rostral pole of the BIN, which lies ventrally at the border between the midbrain and the thalamus (Fig. 1), it is also possible that neurons showing this type of binaural interaction are not found in the BIN.

Our finding that the BIN contains a weakly topographic representation of sound azimuth, which is conveyed by a spatially ordered projection to the SC, suggests similarities with the way in which the map of auditory space in the ICX is projected to the optic tectum in the barn owl (Knudsen and Konishi 1983). Apart from some differences in their internal organization, the two maps in the barn owl midbrain are very similar (Knudsen 1982; Knudsen and Konishi 1978). However, compared with the SC, the ferret BIN contains a rather coarse representation of sound azimuth and no apparent map of elevation. This would suggest that the construction of a precise two-dimensional map of auditory space in the SC relies on converging inputs from neurons in the BIN as well as from other auditory areas. Simultaneous recordings from pairs of units in the BIN support this idea by showing that spikes generated by the unit pair within a 5-ms time window exhibit sharper spatial tuning with bandwidths much closer to those of SC units (King et al. 1997b).

Although the BIN–SC projection is predominantly ipsilateral, the BIN also sends a small projection to rostral parts of the contralateral SC (King et al. 1997a). In addition, our tracing studies reveal a substantial input from the ICX to the SC, although this projection does not appear to be topographically organized. Nevertheless, a map of sound azimuth has been described in this nucleus in the guinea pig (Binns et al. 1992). The role of the ICX in relaying the signals to the SC is uncertain (Binns 1991), and further studies are required to assess the relative contributions of different brain stem and cortical (Meredith and Clemo 1989) inputs to the spatially selective responses of auditory neurons in the SC.

There is now considerable evidence that the development of the auditory space map in the SC is guided by sensory experience (King 1993; Knudsen and Brainard 1995). Based on the consequences of either lesioning one ear or removing the pinnae in infancy, we have proposed previously that the near-threshold monaural map in the SC could potentially act as a template for the subsequent development of sensitivity to binaural cues the values of which correspond to the same region of space (King et al. 1994; Schnupp et al. 1995a). Our present data suggest that the BIN may be more concerned with processing binaural cues for sound location, raising the possibility that the inputs from this nucleus are refined during development as a result of the sensitivity of SC neurons to monaural spectral cues, which presumably arises from a different brain stem pathway.
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