THE JOURNAL OF COMPARATIVE NEUROLOGY 238:65-76 (1985)

Inferior Colliculus of the House Mouse. I. A Quantitative Study of Tonotopic Organization, Frequency Representation, and Tone-Threshold Distribution

IMME STIEBLER AND GÜNTER EHRET
Fakultät für Biologie, Universität Konstanz, D-7750 Konstanz, Federal Republic of Germany

ABSTRACT

Electrophysiological mapping was used to study frequency representation in the inferior colliculus (IC) of the mouse. In the lateral nucleus (LN) only part of the frequency range of hearing was represented and tonotopicity was separate from that in the rest of the IC. Highest frequencies occupied the medial part (M) of the central nucleus (CN). A single complete representation of the hearing range was present only if representations in the dorsal cortex (plus dorsomedial nucleus) and CN (including M) were combined. Continuous isofrequency planes making up these nuclei (without the lateral part of the CN) were reconstructed. They tilted from medial to lateral and from caudal to rostral. The steepness of the slopes increased from caudal to rostral and from dorsal to ventral (i.e., with increasing frequency). Isofrequency planes had similar angles of deviation from the horizontal plane as described for dendritic laminae in the CN. Differences of mapping in the lateral part of the CN from that in the rest of the CN could be explained by the different organization of laminae in this part.

The relative amounts of IC depth and volume occupied by parts of the mouse audible frequency range were quantified. Frequency representation along IC depth was not proportional to that along cochlear length. Compared with the relative density of afferent nerve fiber supply within given frequency ranges represented along the basilar membrane, there is a relative under-representation in the IC up to 15–20 kHz and an over-representation of higher frequencies.

Highest absolute tone sensitivity (lowest threshold) was found in neurons forming a column (running perpendicular to isofrequency planes) in the center of the IC.

Results are discussed with regard to frequency representation, intrinsic neuronal organization, and functional segregation in the IC of mammals.

Key words: frequency-sensitivity mapping, spatial representation, electrophysiology, midbrain

The auditory midbrain center, the inferior colliculus (IC) of mammals, has direct ascending connections from more than ten brainstem nuclei (e.g., Beyerl, '78; Roth et al., '78; Brunso-Bechtold et al., '81; Schweizer, '81; Willard and Ryugo, '83). It is still unclear whether all these afferents project to a single set of tonotopically arranged isofrequency planes including the dorsal cortex (DC), dorsomedial nucleus (DM), central nucleus (CN), and lateral (or external) nucleus (LN) (e.g., Fitzpatrick, '75; Webster et al., '84; Servière et al., '84) or whether there are separate tonotopic representations in the DC, CN, and LN, as other work suggests (Rose et al., '63; Merzenich and Reid, '74; Kitzes,'84). The knowledge of how the frequency representation within the cochlea is transformed into the three-dimensional neural substrate of the IC is of particular im-

Accepted March 29, 1985.

© 1985 ALAN R. LISS, INC.

portance with regard to the projection patterns of the brainstem nuclei. There is evidence for a segregation of major projections from different brainstem nuclei to different places on the isofrequency planes, on the one hand, and for a highly divergent distribution of input from given nuclei on the other (Roth et al., '78; Brunso-Bechtold et al., '81; Oliver, '84; Whitley and Henkel, '84). Thus, from a functional point of view, neural response specialization in the brainstem nuclei might be preserved by projections being restricted to certain areas of the IC and new functional properties might emerge on the substrate of the IC from interactions of divergent projections from different nuclei.

In the present study, we analyze the tonotopicity in four nuclei (DC, DM, CN, LN) of the IC of the mouse *(Mus musculus),* which has become an important animal model for studying auditory function especially with regard to development, pathology, and genetic effects (summarized in Willott, '83). In addition, we present a quantitative analysis of the relative amounts occupied by certain frequency ranges in the IC compared with the frequency representation and nerve fiber density along the basilar membrane of the cochlea. This analysis will show whether there is an over- or under-representation of certain frequency ranges in the IC compared with what can be expected from the cochlear output. Finally, a mapping of tone response thresholds will show whether a functional segregation within isofrequency planes according to sensitivity is present in the IC.

METHODS

Subjects

Young adult house mice *(Mus musculus,* outbred strain NMRI, aged 8–15 weeks) of both sexes from our own colony were used. Only animals that showed responses to frequencies of 50 kHz and higher were analyzed in the electrophysiological mapping study (three females, seven males).

Frequency and threshold mapping

Animals were anesthesized with sodium pentobarbital (Nembutal, 70 mg/kg, intraperitoneally) for surgery. The skin over the skull and the dura overlying the right IC were carefully removed. The animals were rigidly fixed in a headholder (a clamp between the palate and nasal/frontal bones) and placed on a feedback-controlled heating pad (rectal temperature was kept at 38 ± 1°C) in a soundproof and anechoic room. The headholder was adjusted to keep the dorsal surface of the skull in a horizontal position. Steel electrodes were placed stereotactically, with reference to the λ-point of the skull, on the dorsal surface of the IC and were advanced into the IC vertically to the dorsal surface with a remote-controlled hydraulic microdrive (Trent Wells Mark III). Electrode positions were selected so that several penetrations were in a mediolateral line at a certain rostrocaudal coordinate, and in a rostrocaudal line at a certain mediolateral coordinate. Several electrode tracks were in the same plane in frontal or parasagittal sections of the IC. Penetrations were spaced in order to sample the IC in a representative way.

Tones of known frequency (Kontron counter/timer 6001) were produced by a generator (Wavetek 132), shaped into bursts of 70-msec duration (including 10-msec rise and fall times, respectively) and 350-msec interburst intervals, and sent through two attenuaters (Hewlett-Packard 350D) and amplifiers (Hewlett-Packard 466A and Exact 170) to a dynamic speaker for frequencies below 12 kHz and to an electrostatic speaker (Machmerth et al., '75) for higher frequencies. Sounds were delivered freefield to the left ear. The distance between the speakers and the animal was 0.4 m. Sound pressure levels (re. 20 μPa) of the tones were measured at the place of the animals' pinnae by a calibrated 6.35-mm microphone (Bruel and Kjaer 4135) and measuring amplifier (Bruel and Kjaer 2606).

In 108 electrode penetrations we recorded multi- or single-unit responses to tone bursts at 799 predetermined depths within the IC. At each tip position of the electrode the characteristic frequency (CF) and tone response threshold were determined audiovisually as that frequency and intensity at which a response (mostly with a phasic component) became just noticeable. Since spontaneous activity was usually below 3 spikes/second (Ehret and Moffat, '85) and background noise below 0.01 mV this is a straightforward procedure. For construction of electrode tracks, an electrolytic lesion was made at the end of every penetration or at the point of the highest frequency encountered.

Data were obtained only while the animals were in a stable physiological state with heart rate at 8.5/second and breathing rate at 1.9/second. During the recording session the anesthetic state was maintained by a combined injection of Nembutal (10 mg/kg) and chlorprothixene (Taractan, 2.5 mg/kg) as described in Ehret and Moffat ('85).

At the end of the recording session the animals were killed by a Nembutal overdose. The brains were immediately removed and quickly frozen without previous fixation with a freezing microtome (Reichert-Jung, 2700 Frigocut). Serial sections of 20 μm were made and stained with cresyl-violet. The sections were used to determine the nucleus regions in the IC and the electrode tracks and lesions. All tracks and therefore all recording sites could be clearly identified. Figure 1 shows the histology of reconstruction of electrode tracks and recording sites for one mouse as an example.

RESULTS

From our histology, and in comparison with other studies on the IC of the mouse (Ryugo et al., '81; Willard and Ryugo, '83), we distinguished the following areas (compare Fig. 1): (1) The central nucleus (CN; Willard and Ryugo, '83), which composes the central, lateral and ventrolateral part of the central nucleus (Morest and Oliver, '84).

Abbreviations

c	caudal
CN	central nucleus
d	dorsal
DC	dorsal cortex
DM	dorsomedial nucleus
IC	inferior colliculus
LL	lateral lemniscus
LN	lateral nucleus
M	medial part of the CN
r	rostral
v	ventral

Fig. 1. Histology and frequency mapping in mouse CI93. Altogether ten penetrations with marked depths of frequency measurements and lesion sites are visible in three frontal sections through the rostral (A), central (B), and caudal (C) parts of the IC. The nuclei of the IC, lesions, and measured CFs are shown in the camera lucida drawings at the right side of the figure. Dashed lines indicate borders between nuclei that could not unequivocally be determined.

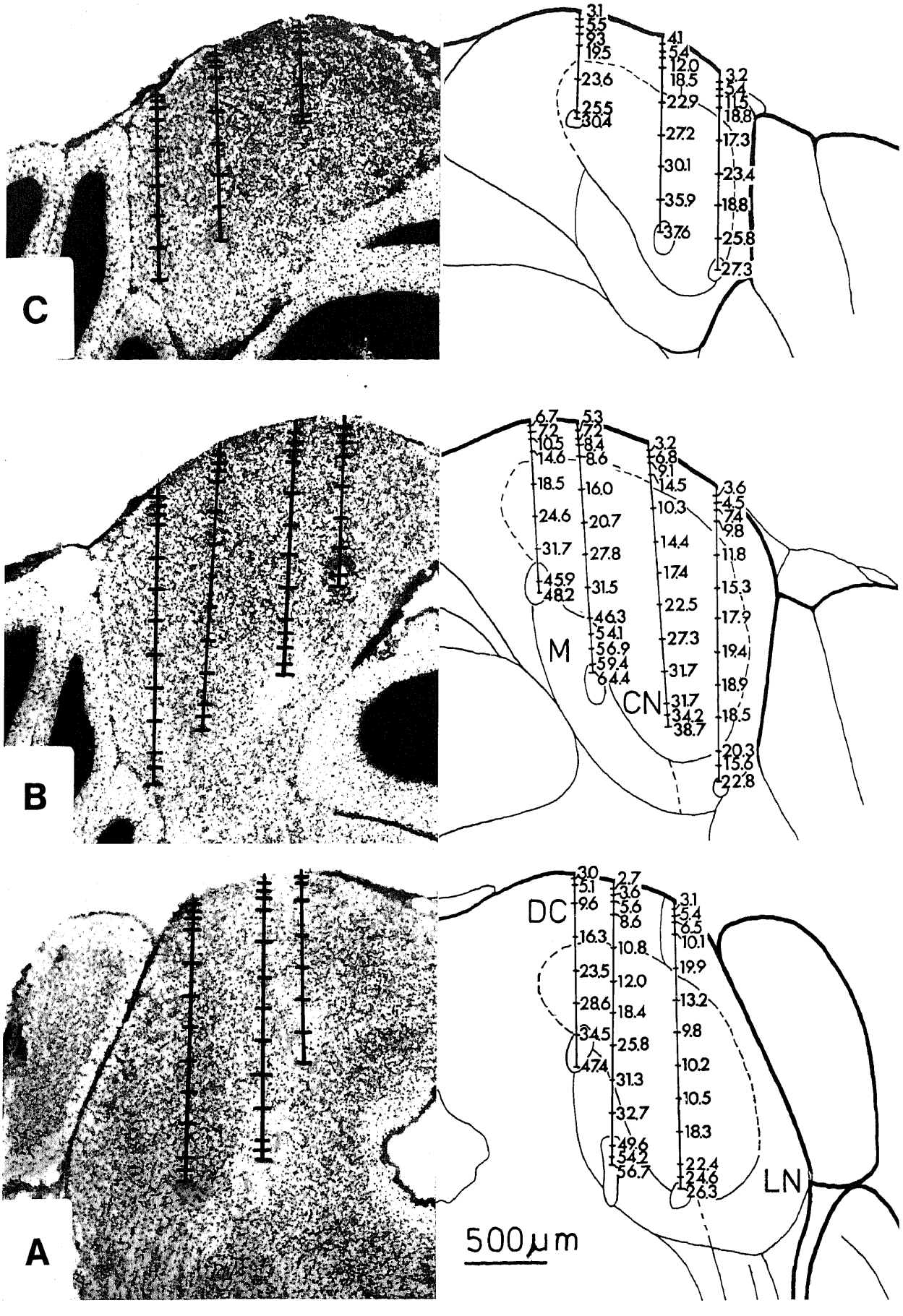

C
3.1
5.5
9.3
19.5
23.6
25.5
30.4
4.1
5.4
12.0
18.5
22.9
27.2
30.1
35.9
37.6
3.2
5.4
11.5
18.8
17.3
23.4
18.8
25.8
27.3
B
6.7
7.2
10.5
14.6
18.5
24.6
31.7
45.9
48.2
5.3
7.2
8.4
8.6
16.0
20.7
27.8
31.5
46.3
54.1
56.9
59.4
64.4
3.2
6.8
9.1
14.5
10.3
14.4
17.4
22.5
27.3
31.7
31.7
34.2
38.7
3.6
4.5
7.4
9.8
11.8
15.3
17.9
19.4
18.9
18.5
20.3
15.6
22.8
M
CN
A
3.0
5.1
9.6
16.3
23.5
28.6
34.5
47.4
2.7
3.6
5.6
8.6
10.8
12.0
18.4
25.8
31.3
32.7
49.6
54.2
56.7
3.1
5.4
6.5
10.1
19.9
13.2
9.8
10.2
10.5
18.3
22.4
24.6
26.3
DC
LN
500μm

Figure 1

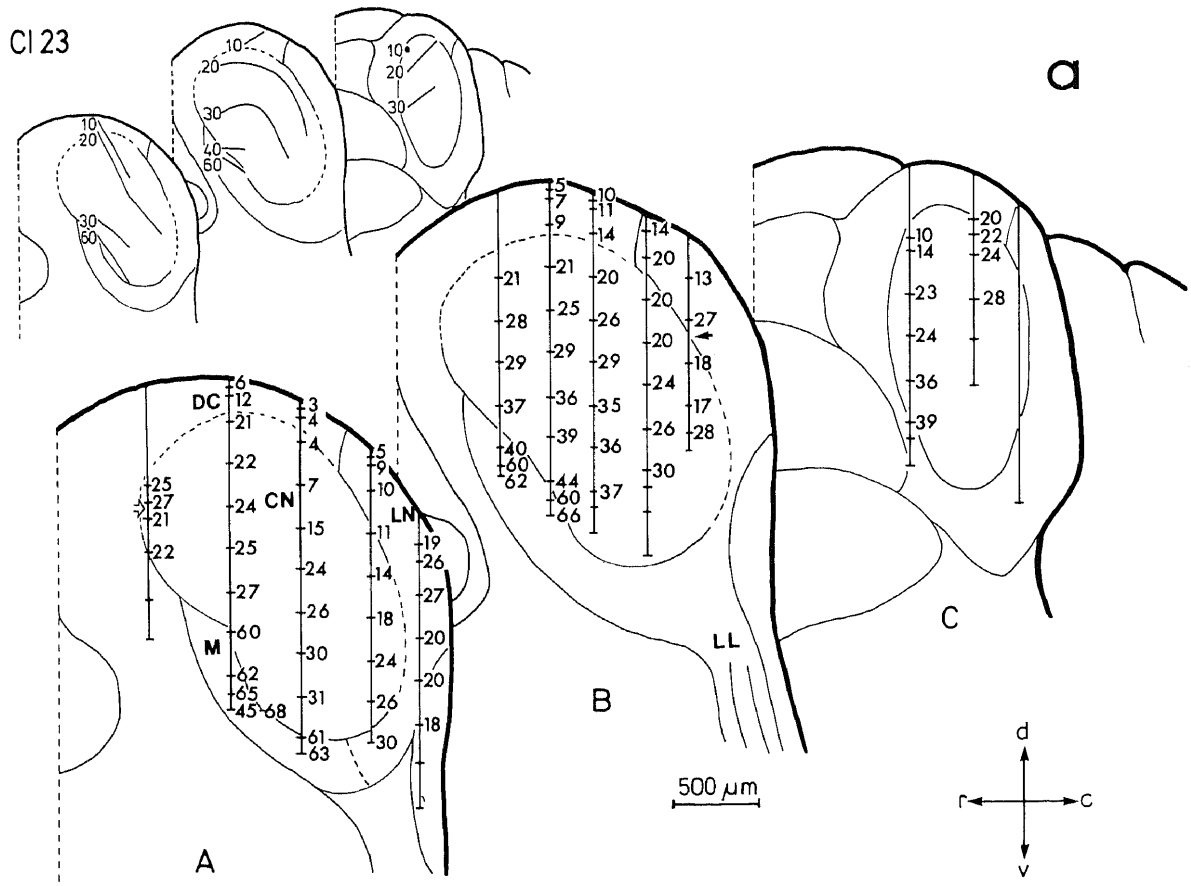


Fig. 2. Camera lucida drawings of frontal sections showing frequency mapping in five of the animals as examples. Sectioning plane A is always the most rostral one. Except for CI35 (700 µm), all A-planes are 800 µm caudal of the λ-point of the skull. The other planes are always 200 µm (CI39, CI41) or 300 µm (others) further caudal from the preceding planes. Black arrows indicate discontinuities of frequency representation at the CN-LN border, white arrows (CI23, CI39) point at irregularities at the most medial border of the CN. The insets show for each plane a reconstruction of isofrequency lines for 10, 20, 30, 40, 50 and 60 kHz.

(2) The medial part (M) of the central nucleus (Morest and Oliver, '84), which is equal to the medial aspect of the external nucleus (Willard and Ryugo, '83).

(3) The lateral nucleus (LN; Oliver and Morest, '84), which corresponds to the lateral part of the external nucleus (Willard and Ryugo, '83).

We were not always able to distinguish unequivocally between the dorsal cortex (DC), the dorsomedial nucleus (DM), and the central nucleus from the cresyl-violet histology. In addition, no irregularities of frequency representation occurred at the borders of DC, DM, and CN in our mapping study (see below). Therefore, we analyzed the DC and DM together with the CN.

Tonotopicity and frequency representation

In Figure 2 complete frequency mappings are shown for five animals as examples. They agree with the data of the remaining five animals and demonstrate the following:

(1) The absolute lowest CF measured in all animals was 2.3 kHz, the highest 68.5 kHz (averaged lowest and highest CFs were 2.8 ± 0.3 kHz and 61.3 ± 5.6 kHz, respectively).

(2) A steady frequency increase with increasing penetration depth was present in all but eight tracks starting in the DC. Responses to tone bursts were absent in five tracks or showed irregularities in three tracks, all situated closest to the midline and outside or at the border of the CN (including the DM) (e.g., Fig. 2b:C; 2c:A; 2d:B,C,D; 2a:A open arrow; 2c:C open arrow). We never found a frequency reversal or discontinuity when passing from the DC to the CN.

(3) In 17 of 32 penetrations starting in the LN, CFs first increased to the border of the CN, then dropped to a lower value in the CN and increased again (e.g., black arrows in Fig. 2a:B; 2b:B; 2c:A,C; 2d:B,C; 2e:A,B,C).

(4) A steady increase with depth in the lateral part of the CN did not occur in 20 of 27 penetrations through this area. The frequency could stay rather constant over a considerable depth (e.g., Fig. 2a:B;, 2b:A,B; 2d:D) or could show irregular changes (e.g., Fig. 2a:B; 2b:C; 2c:A,B; 2e:A,C).

(5) In 13 of 19 penetrations which left the CN at the ventrolateral border to the LN, a sometimes marked drop of CF was seen (e.g., black arrows in Fig. 2b:B; 2c:A,B; 2d:B,D; 2e:B). If the electrode track continued downward into the LN, the CF often increased again.

(6) The highest frequencies were always represented in the medial part (M) of the central nucleus (Fig. 2a–e).

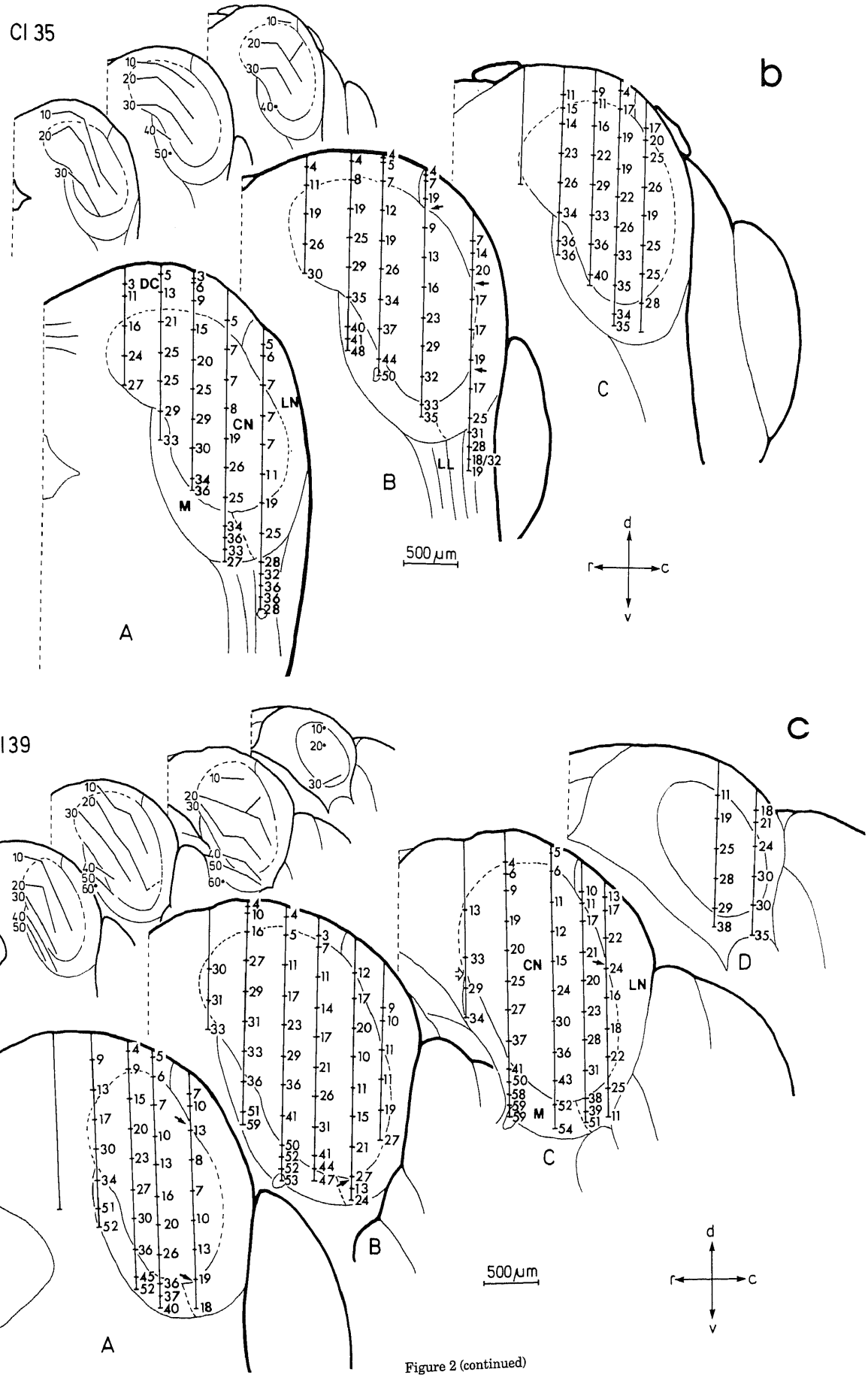
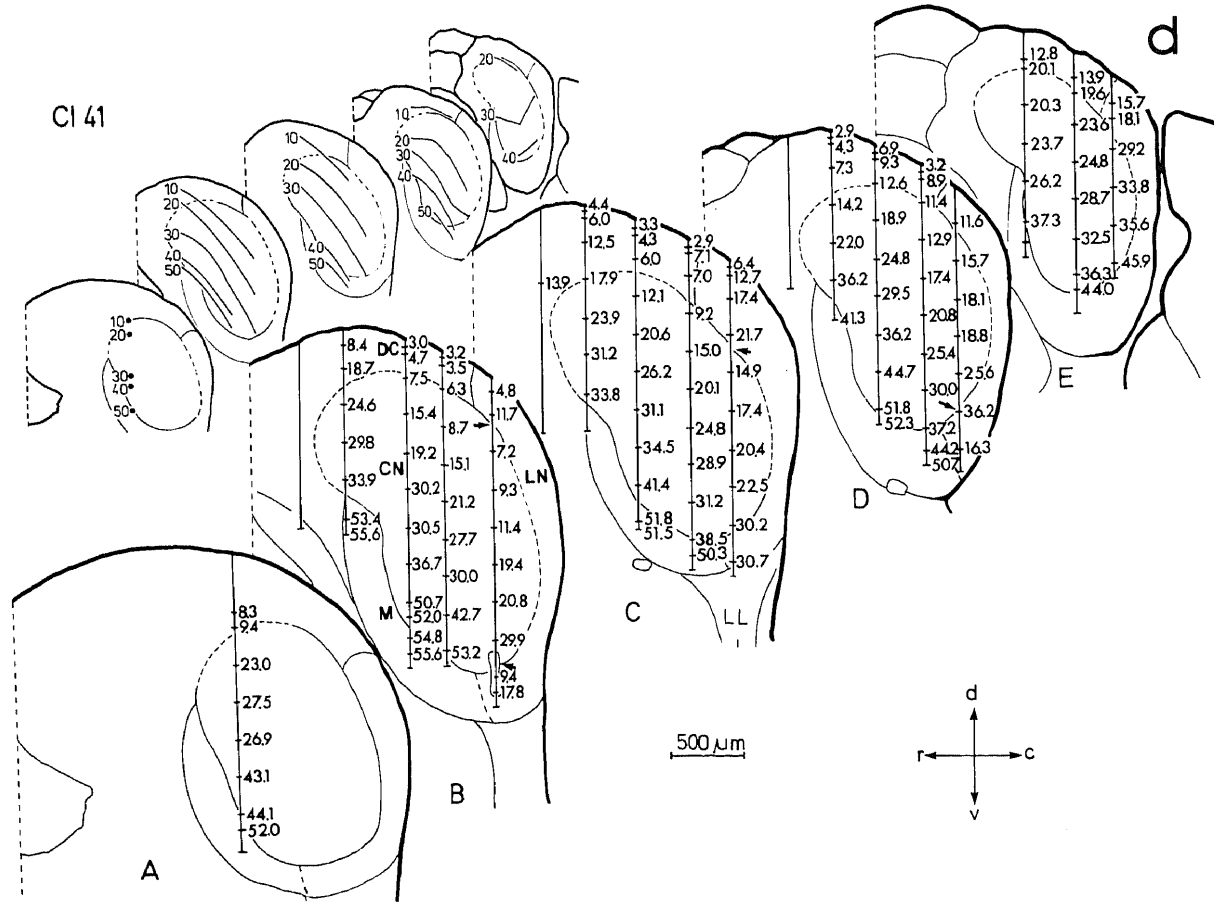


Figure 2 (continued)



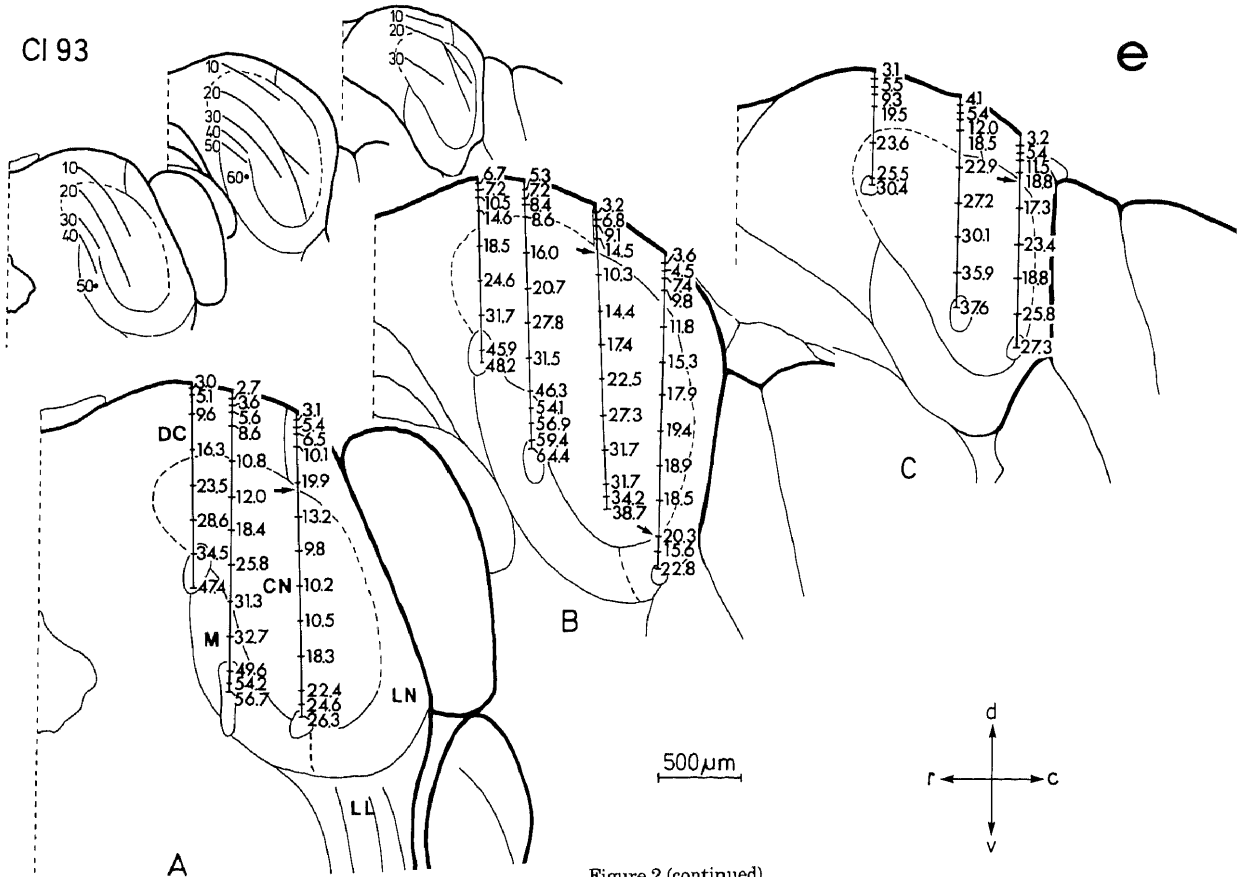


Figure 2 (continued)

(7) At the transition of the CN to the M often a sudden increase of CF for more than 10 kHz over a comparably small increase of depth occurred (e.g., Fig. 2a:A,B; 2c:A,B; 2d:B,C; 2e:A,B).

(8) Penetrations into the periaquaeductal gray and interstitial nucleus of the IC (Willard and Ryugo, '83) or central gray and commissural nucleus (Morest and Oliver, '84) did not reveal any responses to tone bursts (e.g., Fig. 2a:A; 2b:C; 2c:A; 2d:B,C.D.)

The frequency mapping demonstrated in the examples of Figure 2 allowed the construction of isofrequency lines which connect points of the same CFs in different penetrations within the same plane in the IC. Continuous isofrequency lines could be drawn from the DC and DM through the CN and M (insets in Fig. 2a–e). In the lateral part of the CN 10- and 20-kHz isofrequency lines could show an upward bending or a splitting into two branches—one following the steep negative slope ventralward, the other running with a more shallow or even positive slope toward the LN (Fig. 2b:C; 2c:B,C; 2e:B,C). In all except one case of bending or branching of isofrequency lines, lowest thresholds occurred at the most ventral points of representation. We quantified the course of the isofrequency lines (not including the LN), calculated the mean slopes (between the most medial and the most ventral points) and averaged these slopes across animals separately for 10, 20, 30, 40, and 50 kHz and separately for the rostral, central, and caudal thirds of the IC. Table 1 shows the results. The absolute slope values decreased from rostral to caudal for all frequencies (U-test, always $P < .01$). All isofrequency lines except some in the caudal part (e.g., Fig. 2a:C; 2c:D; 2d:E) had negative slopes. Table 1 also indicates an increase of absolute values with increasing frequencies. This relationship is shown in Figure 3 for 10-, 20-, 30-, 40-, 50-, and 60-kHz lines from all positions in the IC. The calculated regression with the negative slope is statistically highly significant ($P < .001$). Thus it is evident that isofrequency lines become steeper from caudal to rostral and from dorsal to ventral, i.e., with increasing frequency.

The total frequency range represented in the different parts of the IC differed markedly. Table 2 shows that there was little variance among animals for the lowest and highest frequencies found in each of the distinguished nucleus areas. The CN + DC + DM made up the largest frequency range from the lowest to rather high frequencies. The M part of the IC contained only high frequencies up to the upper frequency limit. In the LN, lowest frequencies and the total high-frequency range were absent. Thus a complete representation of the whole audible frequency range of the mouse was obtained only if the DC, DM, CN, and M were all considered together.

TABLE 1. Average Slopes and Angles From the Horizontal Plane of Isofrequency lines for the Rostral, Central, and Caudal Thirds and for the Whole IC: Data From All Animals Included

Frequency (kHz)	Rostral		Central		Caudal		Whole IC	
	Slope	Angle	Slope	Angle	Slope	Angle	Slope	Angle
10	−1.31	53°	−0.34	19°	−0.13	7°	−0.56	29°
20	−1.41	55°	−1.02	46°	−0.37	20°	−0.93	43°
30	−1.30	52°	−0.97	44°	−0.35	19°	−0.85	40°
40	−1.65	59°	−1.08	47°	—	—	−1.11	48°
50	−1.57	58°	−1.29	52°	—	—	−1.36	54°
60	—	—	−1.03	46°	—	—	−1.12	48°

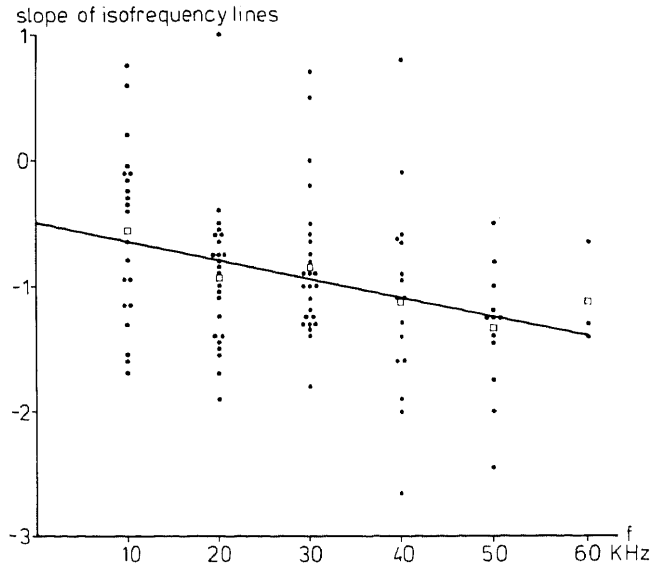


Fig. 3. Average slopes of isofrequency lines from all animals plotted against the frequencies. The regression line indicates a significant ($P < .001$) relation between slope and CF. The open squares are the means of the slopes at each frequency indicated.

TABLE 2. Frequency Ranges (kHz) Represented in the Different Nucleus Areas in All Animals

Animal	DC + DM + CN	M	LN
CI19	3–50	40–62	7–24
CI23	3–44	39–68	5–27
CI27	3–39	31–60	5–30
CI35	3–40	33–50	4–28
CI39	3–44	36–59	7–35
CI41	2.9–53.2	37.2–55.6	4.8–30.7
CI51	3.9–36.0	26.6–64.0	8.4–21.2
CI55	2.3–58.6	40.8–68.5	4.4–24.9
CI70	2.4–37.6	45.7–61.8	2.7–22.7
CI93	2.7–46.3	32.7–64.4	3.1–22.8

Since our relative sample sizes of the different IC areas were similar in all animals investigated, and CFs were always determined at comparable depths, we were able to calculate the relative representation of frequency ranges over all three dimensions of the IC. We estimated the CFs at every 250-μm depth in each penetration starting with the first and ending with the last real frequency value measured and summing up the occurrence rates in frequency classes which corresponded to those covering every 0.5-mm length of the basilar membrane of this mouse strain (after Ehret, '75; $f = 3{,}350\ (10^{0.21x} - 1)$, with x = distance from apex). Figure 4 shows the percent frequency representation separately for the CN (including DC and DM), for the medial part (M) of the CN, for the LN, and for the whole IC. Clearly, the frequency range between 15 and 34 kHz was strongly represented in the volume occupied by the DC + DM + CN while lower and higher frequencies were less common. Frequencies above 45 kHz were almost exclusively represented in M. The LN contained frequencies between 3.6 and 15 kHz and between 26 and 34 kHz in

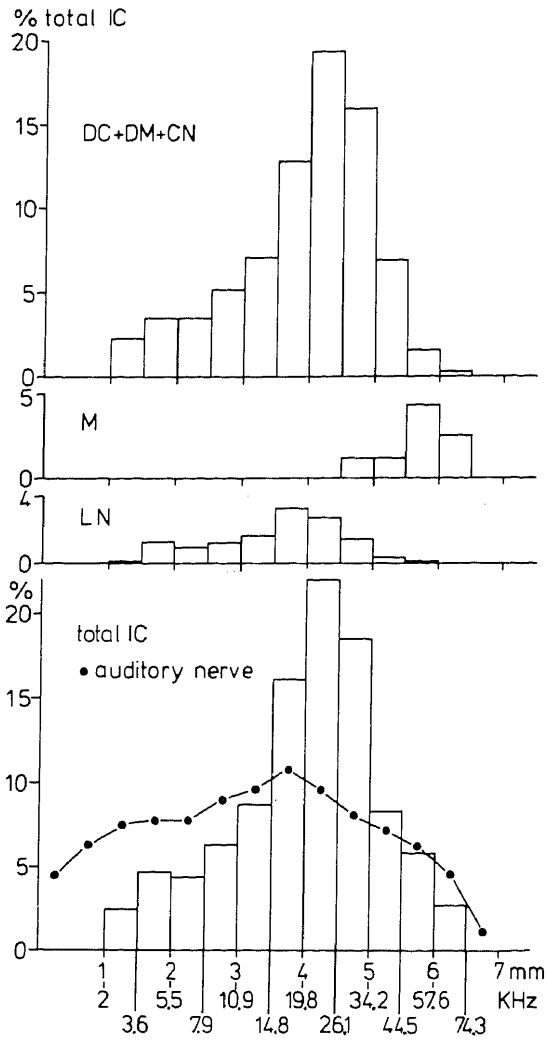


Fig. 4. Histograms of the percent volume occupied in the IC by certain frequency ranges, all corresponding to 0.5-mm lengths of the basilar membrane. Separate histograms are presented for the DC + DM + CN (without M), for the LN, M, and the total IC. The relative numbers of afferent fibers of the auditory nerve innervating given cochlear section are also shown (from Ehret, '79).

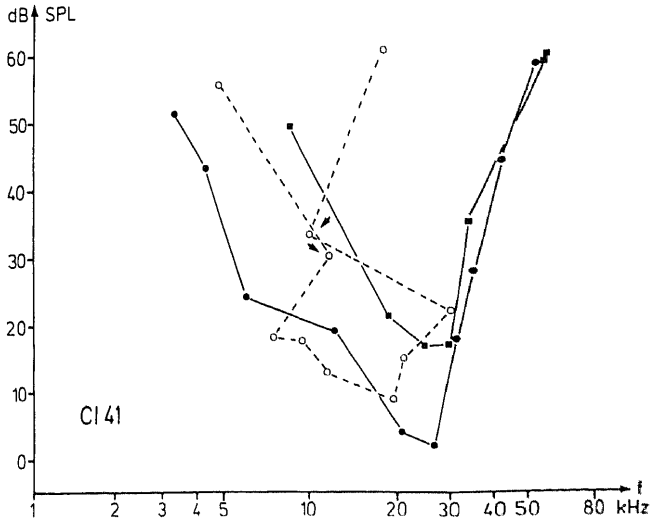


Fig. 5. Example of tone response thresholds in three penetrations through the medial (squares), central (closed circles), and lateral (open circles) parts of the IC of mouse CI41. The arrows indicate discontinuities when passing from the lateral to the central and back to the lateral nucleus.

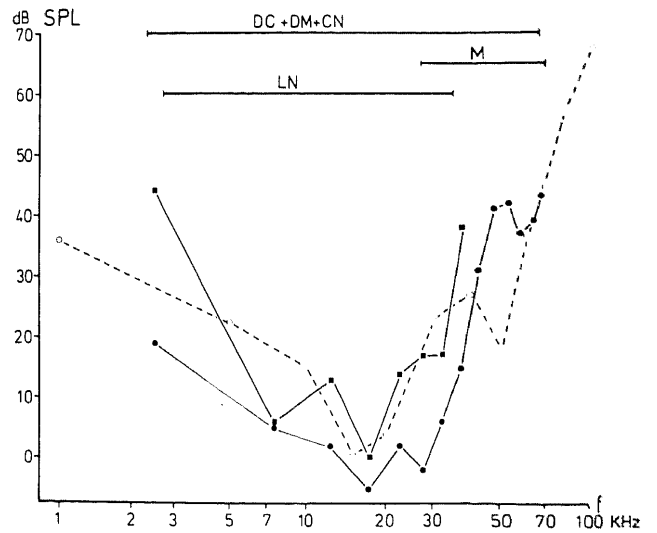


Fig. 6. Lowest tone thresholds measured within 5-kHz classes plotted against frequency. Closed circles, values from DC + DM + CN; squares, values from LN; open circles, behavioral thresholds. The maximum frequency ranges covered by the DC + DM + CN, by the LN, and by the M are also indicated.

similar proportions and had a peak of representation between 15 and 26 kHz. All together, 56% of the whole IC volume represented little more than one octave of CF (15–34 kHz) or a frequency range covering only 22% of the basilar membrane.

Tone-threshold distribution

The threshold sensitivities of neural tone burst responses depend on the frequencies of the tones. This becomes evident in Figure 5, in which tone response thresholds from three typical penetrations are plotted as a function of CF. These examples show that an electrode track through the central part of the IC revealed lower thresholds than if placed closer to the midline or including the lateral nucleus. Thresholds in the lateral track were high when related to the LN. After the electrode passed into the CN, which could be noticed by the sudden drop of CF, threshold values decreased markedly. When the electrode left the CN, the frequency dropped to a lower value (arrow) in the LN, which was again accompanied by a threshold increase.

The differences between the lowest thresholds in the LN and CN are also obvious in Figure 6. Here the lowest thresholds measured in our sample are plotted as a function of CF (within 5-kHz classes). In all CF classes thresholds in the LN were higher than in the rest of the IC although the

lowest thresholds always occurred between 15 and 20 kHz. Thresholds contributed by the medial part (M) of the CN showed a small relative sensitivity maximum near 55 kHz.

A summary of the threshold data of all animals is presented in Figure 7. We plotted the locations of the lowest thresholds in each 5-kHz CF class from each mouse on idealized frontal sections through the rostral, central, and caudal parts of the IC. Significantly more lowest thresholds were situated in the central compared with the rostral and caudal parts (χ^2 test, $P < .001$), and significantly more in the central aspect of the central part compared with the medial and lateral aspects ($P < .001$). In the LN there was only one case of lowest threshold in any frequency class. We can conclude, therefore, that lowest tone thresholds or highest tone response sensitivities are concentrated in a column in the core of the IC. This column includes parts of the DC, CN, and M.

DISCUSSION

Tonotopicity and frequency representation

The present mapping revealed a single tonotopically arranged set of isofrequency planes making up all of the dorsal cortex and dorsomedial nucleus, the central nucleus except its lateral part, and the medial part of the central nucleus (which is the medial aspect of the external nucleus after a different nomenclature). Except for the caudal third of the IC, frequencies within this area increased from dorsal and lateral to ventral and medial. The isofrequency planes clearly crossed anatomical borders of DC, DM, and CN without discontinuities or irregularities. These data extend and substantiate earlier findings on the mouse IC (Harnischfeger, '78; Willott and Urban, '78; Willott, '84). They are in agreement with frequency mappings of the IC of the rat (Clopton and Winfield, '73), rabbit (Aitkin et al., '72), and cat (Semple and Aitkin, '79). They also support the observations made in cats (Servière et al., '84) and monkeys (Fitzpatrick, '75; Webster et al., '84) that isofrequency planes extend into the DC and DM areas of the IC. We could not find, however, discontinuities or frequency reversals when passing through the layers of the DC or from the DC into the CN, which is a common feature in cats (Merzenich and Reid, '74; Servière et al., '84). It could be that the superficial layers of the DC giving rise to a discontinuity of frequency increase are very thin in the mouse, so that we could not map this area.

A discontinuity consistently appeared in the mouse when passing from the lateral (external) into the central nucleus. CFs increased in the LN from lateral to medial and from dorsal to ventral (Fig. 2) and dropped at the border to the CN. Our data thus agree with data from possum (Aitkin et al., '78), rat (Clopton and Winfield, '73), gerbil (Kitzes, '84), and cat (Rose et al., '63), all of which suggest a frequency representation in the LN independent of that in the rest of the IC. From our data a tonotopic arrangement in the LN can be inferred with low frequencies represented laterally and dorsally and high frequencies at the border to the CN (medially) and ventrally. This tonotopic arrangement and the absence of representation of high ultrasonic frequencies has to be considered when the functions of this nucleus are discussed in view of its substantial somatosensory and motor connections (e.g., RoBards et al., '76; Aitkin et al., '81). The frequency range represented in the LN covers the spectra of mouse pup distress calls and of adult female defensive calls, both of which release well-defined reflexive motor responses in the receivers (e.g., Haack et al., '83), and of mouse pup wriggling calls (Ehret and Bernecker,

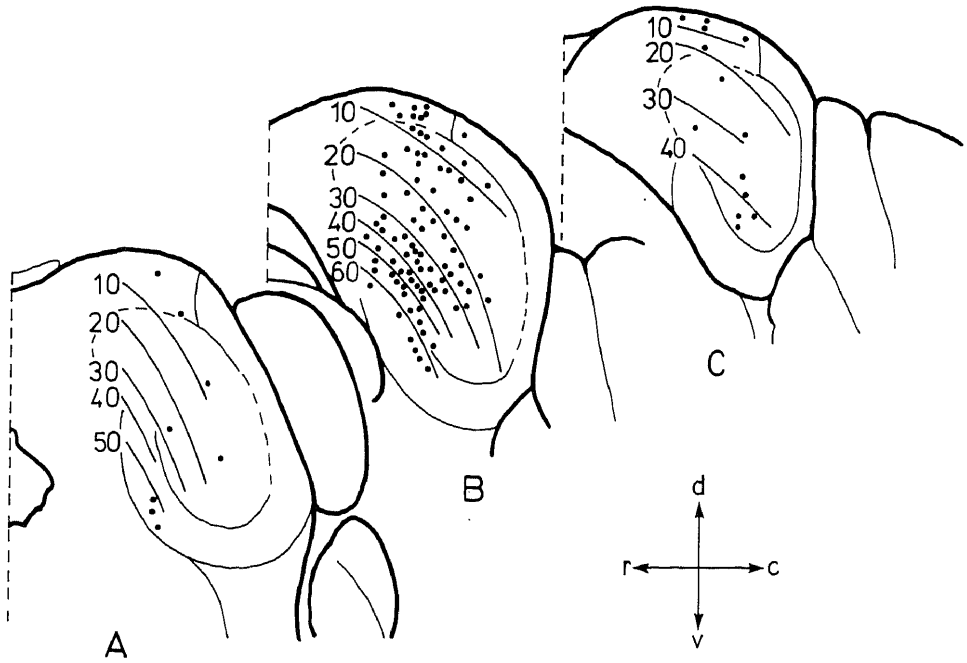


Fig. 7. Idealized frontal sections through the rostral (A), central (B), and caudal (C) parts of the IC showing isofrequency lines and the locations of the lowest thresholds in each 5-kHz class from each mouse. Lowest thresholds are concentrated in the center of the IC in a mediolateral as well as in a rostrocaudal orientation.

'85), which release instinctive maternal behavior. In the first case, a calling pup being carried by an adult mouse is dropped immediately, in the second case an investigating male is stopped rather abruptly, and in the third case a short-latency licking response is most frequently elicited in the mother. If, as suggested by Aitkin et al. ('81), the LN is a center of acousticomotor integration, it may play a significant role in the mediation of the above-mentioned sound-evoked behaviors. In addition, wriggling sound-evoked licking of mouse pups is enhanced if the mother receives simultaneous somatosensory stimulation of moving pups, indicating heterogeneous summation of somatosensory and auditory input which could take place in the LN of the IC.

An interesting aspect arises from a comparison of the present mouse data with the frequency representation in the IC of the echolocating horseshoe bat *(Rhinolophus ferrumequinum)*. Most neurons tuned to the constant-frequency part of the echolocation signal (the filter region: 77–84 kHz) are located ventromedially over a depth of about 600 μm and are separated from the rest of the IC by a layer of tone-insensitive neurons (Pollak and Schuller, '81). This ventromedial filter region of the bat IC has much in common with the M part of the IC of the mouse. In both areas CFs change little with penetration depth and both cover only a small frequency range at the upper frequency limit of hearing. Instead of a tone-insensitive layer of neurons between CN and M, we often found a sudden frequency increase when passing from the CN into its medial part. Another similarity between bat and mouse arises from the function the neurons in the M part of the IC are involved in. In the bat the M neurons are tuned to the highly important echolocation frequencies; in the mouse they process ultrasounds of mouse pups which are used by adults to locate a lost pup (e.g., Haack et al., '83).

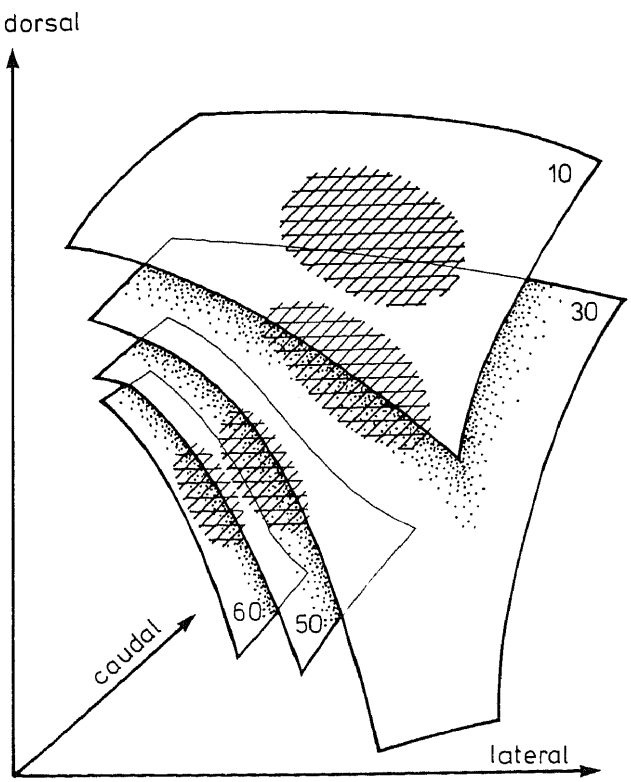


Fig. 8. A reconstruction of isofrequency planes in the IC of the mouse. The planes do not include the lateral part of the CN and the LN. The crosshatched areas indicate zones where neurons with the lowest tone response thresholds are concentrated. These zones form a column penetrating the centers of the isofrequency planes.

Isofrequency planes could be arranged within the three dimensions of the IC. Figure 8 shows a schematic reconstruction of isofrequency planes which make up the DC, DM, CN (without its lateral part), and M. The isofrequency planes show a tilt from medial to lateral and from caudal to rostral with the steepness of this tilt depending on the given frequencies. These contours are comparable in shape to those described for the IC of cats (Semple and Aitkin, '79; Servière et al., '84) and monkeys (Fitzpatrick, '75; Webster et al., '84). The slope values of the isofrequency contours in cats and monkeys also have properties similar to those seen in the mouse. They increase from dorsal to ventral and from caudal to rostral. In cats, the slopes from dorsomedial to ventrolateral range between about 45 and 60° from the horizontal plane (Servière et al., '84), and in the macaque monkey between about 20 and 60° (Webster et al., '84). In the mouse we measured average slopes in the central IC of 19–52° (Table 1). These slope values and the orientation of the isofrequency planes are in agreement with the orientation of dendritic laminae of neurons studies with Golgi methods in the IC of the cat (about 27–57°: Morest and Oliver, '84; about 43–58°: Oliver and Morest, '84) and of the mouse (about 22–53°: Willard and Ryugo, '83). In the central nucleus (including its medial part), the slopes of these laminae increased from dorsal to ventral (except at the ventral border of the IC) and from caudal to rostral, as do the isofrequency contours. We may conclude, therefore, that isofrequency planes in the central nucleus of the IC (including its medial part) basically follow the orientation of the dendritic laminae in the CN. Isofrequency contours, however, do not necessarily depend on the presence of a regularly laminated structure, since they extend into the DC and DM, which apparently have no regular lamination of dendrites (Willard and Ryugo, '83; Morest and Oliver, '84).

The isofrequency planes could not be readily extended into the lateral part of the CN, because a linear frequency increase with penetration depth was often absent (Figs. 1, 2). This finding is consistent with results from other studies. Dorsal to ventral penetrations through the lateral part of the CN did not reveal a steady CF increase in the rat (Clopton and Winfield, '73) and horseshoe bat (Pollak and Schuller, '81). Oliver and Morest ('84) showed that the dendritic laminae in the lateral part of the CN of the cat are orderly in orientation and run almost perpendicular to those in the central part of the CN. This would suggest an upward bending of isofrequency lines in the lateral CN in frontal planes. Such a prediction is supported by half of our data, where in ten of 20 frontal sectioning planes (e.g., Fig. 2b:C; 2c:B,C,D; 2d:E) an upward bending of isofrequency lines or a splitting into two branches, one continuing downward and the other (indicated by dashed lines in the insets of the figures) in an upward direction, are visible. Thus, a considerable body of our data present the first physiological correlate for the orientation of dendritic laminae found in the lateral part of the CN (Oliver and Morest, '84). In conclusion, our physiological frequency mappings support

the distinctions made on anatomical grounds between the central nucleus and its lateral and medial parts (Morest and Oliver, '84).

Comparison with cochlear nerve fiber density

The relative amount of representation in the IC volume of frequency ranges corresponding to 0.5 mm of the basilar membrane is shown in Figure 4 together with the relative density of afferent nerve fibers in the same cochlear sections (from Ehret, '79). An increase of representation with increasing frequency can be seen in the cochlear nerve up to about 20 kHz and in the IC up to about 26 kHz. Then a decrease of representation follows in both cases up to the upper frequency limit. The representation of frequencies up to about 15 kHz is relatively lower in the IC compared with the auditory nerve, while frequencies between about 15 and 44 kHz obviously have a relatively higher representation in the IC.

These comparisons between periphery and IC are based on the geometry only. They do not consider, e.g., the densities of neuronal packing in different parts of the IC, which appear to be higher in the DC than in the CN (Rockel and Jones, '73; Fitzpatrick, '75; Morest and Oliver, '84). Thus the relative under-representation of lower frequencies derived from depth measurements might not be true if numbers of neurons within given depth sections of the IC were compared with peripheral fiber density.

Our data of frequency representation per unit depth basically agree with results from the cat (Merzenich and Reid, '74), which also indicate a deviation from a proprotionality between collicular depth and cochlear length similar to that we found in the mouse.

Representation of tone sensitivity

Tone response threshold curves derived from neurons in the IC resembled the behavioral auditory threshold curve of this mouse strain (Ehret, '74; compare Fig. 6). Since in the LN of the IC only part of the frequency range is represented, we can suggest that the LN is not involved in behavioral auditory threshold assessment, while at least DC and CN (including M) should be considered.

We found that neurons with highest sensitivities (lowest thresholds) were concentrated in a column in the center of the IC (Figs. 7, 8). This column extends perpendicular to the isofrequency planes from the DC through the CN and M and presents the first physiological segregation described for the IC concerning sensitivity. Neurons with certain binaural properties have already been shown to be concentrated in different areas of the central nucleus of the IC of the cat (Roth et al., '78; Semple and Aitkin, '79). These findings of physiological, probably functional, segregations of neuronal populations in the IC are important on the background of differential projections from the auditory brainstem nuclei to different parts of the IC. Available evidence from studies in cats (Osen, '72; Roth et al., '78; Brunso-Bechtold et al., '81; Oliver, '84) and mice (Ryugo et al., '81; Willard and Ryugo,'83) shows predominant projections from the cochlear nucleus and superior olive complexes to the central part of the IC. Physiological studies will have to show whether and how all these projections participate in forming the low-threshold core. One possible arrangement would be that each of the mentioned brainstem nuclei occupied a sector of the low-threshold field in every isofrequency plane (Fig. 8).

ACKNOWLEDGMENTS

This study was supported by the Deutsche Forschungsgemeinschaft, Eh 53/8-5.

LITERATURE CITED

Aitkin, L.M., S. Fryman, D.W. Blake, and W.R. Webster (1972) Responses of neurons in the rabbit inferior colliculus. I. Frequency-specificity and topographic arrangement. Brain Res. 47:77–90.

Aitkin, L.M., B.M.H. Bush, and G.R. Gates (1978) The auditory midbrain of a marsupial: The brush-tailed possum *(Trichosurus vulpecula).* Brain Res. *150:*29–44.

Aitkin, L.M., C.E. Kenyon, and P. Philpott (1981) The representation of the auditory and somatosensory systems in the external nucleus of the cat inferior colliculus. J. Comp. Neurol. *156:*25–40.

Beyerl, B.D. (1978) Afferent projections to the central nucleus of the inferior colliculus in the rat. Brain Res. *145:*209–223.

Brunso-Bechtold, J.K., G.C. Thompson, and R.B. Masterton (1981) HRP study of the organization of auditory afferents ascending to the central nucleus of the inferior colliculus in cat. J. Comp. Neurol. *197:*705–722.

Clopton, B.M., and J.A. Winfield (1973) Tonotopic organization in the inferior colliculus of the rat. Brain Res. *56:*355–358.

Ehret, G. (1974) Age-dependent hearing loss in normal hearing mice. Naturwissenschaften *61:*506.

Ehret, G. (1975) Masked auditory thresholds, critical ratios, and scales of the basilar membrane of the house mouse *(Mus musculus).* J. Comp. Physiol. *103:*329–341.

Ehret, G. (1979) Quantitative analysis of nerve fiber densities in the cochlea of the house mouse *(Mus musculus).* J. Comp. Neurol. *183:*73–88.

Ehret, G., and A.J.M. Moffat (1985) Inferior colliculus of the house mouse: II. Single unit responses to tones, noise and tone-noise combinations as a function of sound intensity. J. Comp. Physiol. (in press).

Ehret, G., and C. Bernecker (1985) Non-ultrasound communication by mouse pups *(Mus musculus):* Wriggling sounds release maternal behavior. Anim. Behav. (Submitted)

Fitzpatrick, K.A. (1975) Cellular architecture and topographic organization of the inferior colliculus of the squirrel monkey. J. Comp. Neurol. *164:*185–208.

Haack, B., H. Markl, and G. Ehret (1983) Sound communication between parents and offspring. In J.F. Willott (ed): The Auditory Psychobiology of the Mouse. Springfield: Thomas, pp. 57–97.

Harnischfeger, G. (1978) Single unit study in the inferior colliculus of the house mouse *(Mus musculus).* Neurosci. Lett. *9:*279–284.

Kitzes, L.M. (1984) Some physiological consequences of neonatal cochlear destruction in the inferior colliculus of the gerbil, *Meriones unguiculatus.* Brian Res. *306:*171–178.

Machmerth, H., D. Theiss, and H.-U. Schnitzler (1975) Konstruktion eines Luftschallgebers mit konstantem Frequenzgang im Bereich von 15 kHz bis 130 kHz. Acustica *34:*81–85.

Merzenich, M.M., and M.D. Reid (1974) Representation of the cochlea in the inferior colliculus of the cat. Brain Res. *77:*397–415.

Morest, D.K., and D.L. Oliver (1984) The neuronal architecture of the inferior colliculus in the cat: Defining the functional anatomy of the auditory midbrain. J. Comp. Neurol. *222:*209–236.

Oliver, D.L. (1984) Dorsal cochlear nucleus projections to the inferior colliculus in the cat: A light and electron microscopic study. J. Comp. Neurol. *224:*155–172.

Oliver, D.L., and D.K. Morest (1984) The central nucleus of the inferior colliculus in the cat. J. Comp. Neurol. *222:*237–264.

Osen, K.K. (1972) Projections of the cochlear nuclei on the inferior colliculus in the cat. J. Comp. Neurol. *144:*355–372.

Pollak, G.D., and G. Schuller (1981) Tonotopic organization and encoding features of single units in inferior colliculus of horseshoe bats: Functional implications for prey identification. J. Neurophysiol. *45:*208–226.

RoBards, M.J., D.W. Watkins, and R.B. Masterton (1976) An anatomical study of some somesthetic afferents to the intercollicular terminal zone of the midbrain of the opossum. J. Comp. Neurol. *170:*499–524.

Rockel, A.J., and E.G. Jones (1973) The neuronal organization of the inferior colliculus of the adult cat I. The central nucleus. J. Comp. Neurol. *147:*11–60.

Rose, J.E., D.D. Greenwood, J.M. Goldberg, and J.E. Hind (1963) Some discharge characteristics of single neurons in the inferior colliculus of

the cat. I. Tonotopical organization, relation of spike-counts to tone intensity, and firing patterns of single elements. J. Neurophysiol. *26:*294–320.

Roth, G.L., L.M. Aitkin, R.A. Andersen, and M.M. Merzenich (1978) Some features of spatial organization of the central nucleus of the inferior colliculus of the cat. J. Comp. Neurol. *182:*661–680.

Ryugo, D.K., F.H. Willard, and D.M. Fekete (1981) Differential afferent projections to the inferior colliculus from the cochlear nucleus in the albino mouse. Brain Res. *210:*342–349.

Schweizer, H. (1981) The connections of the inferior colliculus and the organization of the brainstem auditory system in the greater horseshoe bat *(Rhinolophus ferrumequinum).* J. Comp. Neurol. *201:*25–49.

Semple, M.N., and L.M. Aitkin (1979) Representation of sound frequency and laterality by units in central nucleus of cat inferior colliculus. J. Neurophysiol. *42:*1626–1639.

Servière, J., W.R. Webster and M.B. Calford (1984) Isofrequency labelling revealed by a combined [^{14}C]-2-deoxyglucose, electrophysiological, and horseradish peroxydase study of the inferior colliculus of the cat. J. Comp. Neurol. *228:*463–477.

Webster, W.R., J. Servière, D. Crewther, and S. Crewther (1984) Iso-frequency 2-DG contours in the inferior colliculus of the awake monkey. Exp. Brain Res. *56:*425–437.

Whitley, J.M., and C.K. Henkel (1984) Topographical organization of the inferior collicular projection and other connections of the ventral nucleus of the lateral lemniscus in the cat. J. Comp. Neurol. *229:*257–270.

Willard, F.H., and D.K. Ryugo (1983) Anatomy of the central auditory system. In J.F. Willott (ed): The Auditory Psychobiology of the Mouse. Springfield: Thomas, pp. 201–304.

Willott, J.F. (ed) (1983) The Auditory Psychobiology of the Mouse. Springfield: Thomas.

Willott, J.F. (1984) Changes in frequency representation in the auditory system of mice with age-related hearing impairment. Brain Res. *309:*159–162.

Willott, J.F., and G.P. Urban (1978) Response properties of neurons in nuclei of the mouse inferior colliculus. J. Comp. Physiol. *127:*175–184.