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# Differences in FM response correlate with morphology of neurons in the rat inferior colliculus

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**Summary.** The response characteristics to linear frequency sweeps were studied in two groups of FM (frequency modulation) sensitive neurons in the rat inferior colliculus. 'FM specialized' cells responded to frequency sweeps but not to pure tones. 'Mixed' cells responded to both frequency sweeps and pure tones. FM specialized cells preferred faster and broader sweeps of higher intensity than did mixed cells and were more directionally selective. In addition, FM specialized cells were more sharply tuned to FM velocity and FM range and had longer response latencies. Physiologically identified FM cells stained intracellularly with horseradish peroxidase revealed differences in morphology correlating with the differences in their responses to tones. FM specialized cells had larger dendritic fields, more dendritic branching and more dendritic spines than did mixed cells. The findings are taken as evidence that the two groups of inferior colliculus neurons are both functionally and morphologically distinct.

**Key words:** Frequency modulation – Intracellular recordings – Horseradish peroxidase – Inferior colliculus – Rat

## Introduction

Sensitivity to frequency modulation (FM) has been demonstrated for neurons of the inferior colliculus (IC) in a number of species (Clopton and Winfield 1974; Nelson et al. 1966; Rees and Moller 1983; Suga 1965a, 1968, 1969; Watanabe and Ohgushi 1968). Amongst the diverse FM response selectivities observed for the IC, two types of cells are consistently identified: (1) cells which respond only to tones of changing frequency and are not driven by steady tones; and (2) cells that show a strong preference for the direction of the tone sweep.

The first attempt to systematically classify FM cells of the IC was made in the bat by Suga (1968). He compared the responses of IC neurons to pure tones, FM sweeps and, later, noise bursts. He differentiated three broad categories of cells (FM insensitive, FM sensitive, and asymmetrical) within which nine subcategories of cells were differentiated. Less than 2% of the cells in the IC were specialized to detect FM signals.

Subsequent studies of FM neurons in the auditory system of other animals (Britt and Starr 1976; Erulkar et al. 1968; Rees and Moller 1983; Watanabe and Ohgushi 1968) stressed the response symmetry to tone sweeps (i.e. the relative magnitude of response to the rising and falling phases of FM sweeps, also known as 'directional selectivity'), and FM responses were frequently not predictable from responses to pure tones.

In a recent study in the rat (Poon et al. 1991), a substantial portion (>25%) of the IC neurons were shown to detect FM sounds in a specialized manner akin to that in the cortical cells (Evans and Whitfield 1964; Mendelson et al. 1985; Phillips et al. 1985; Suga 1965a, b, c). A new scheme for classifying IC cells based on their selectivity in response to both FM and pure tones was proposed. Three types of cells were differentiated, 'FM specialized' (representing a specialized form of FM selectivity), 'mixed', and 'FM insensitive' (Table 1, Figs. 1, 2). Two of the three (FM specialized and mixed) were FM sensitive. The response differences between the two types of FM sensitive cells were not examined in detail. In the first part of this study, we examined the classification of

**Table 1.** Distribution of 844 click sensitive IC neurons in the three categories according to their responses to FM and pure tones

| Cell type | Stimulus | | Number of cells (% of total) |
|---|---|---|---|
| | Pure tone | FM tone | |
| FM sensitive FM specialized | – | + | 285 (34%) |
| Mixed | + | + | 365 (43%) |
| FM insensitive | + | – | 194 (23%) |
| Number of cells (% of total) | 559 (66%) | 650 (77%) | 844 (100%) |

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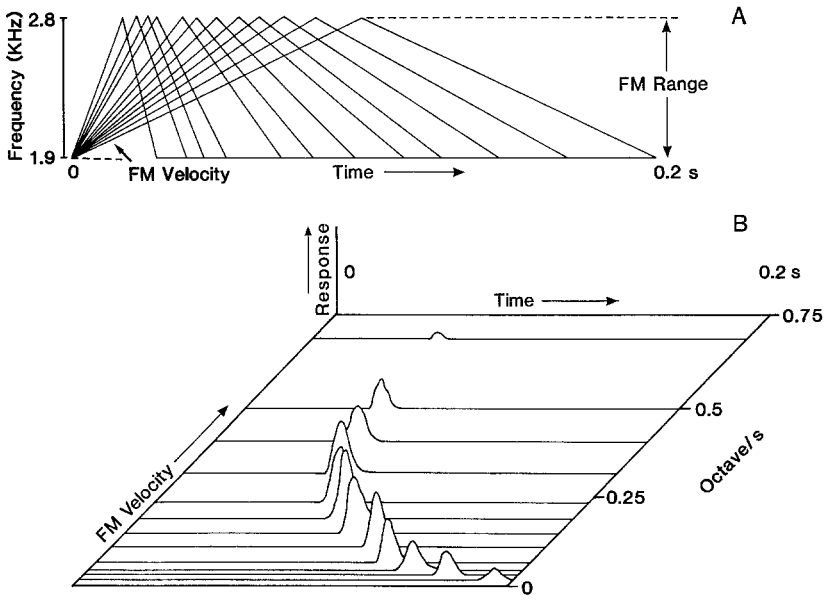


**Fig. 1A–B.** The FM stimulus and an example of a cell response in the IC. **A** Scheme of the modulating signals used to generate the FM stimulus. **B** Response of an FM specialized cell to the FM stimuli shown in **A**; each tracing represents the PSTH to 50 stimuli. This cell responded only to the rising phase of the ramp

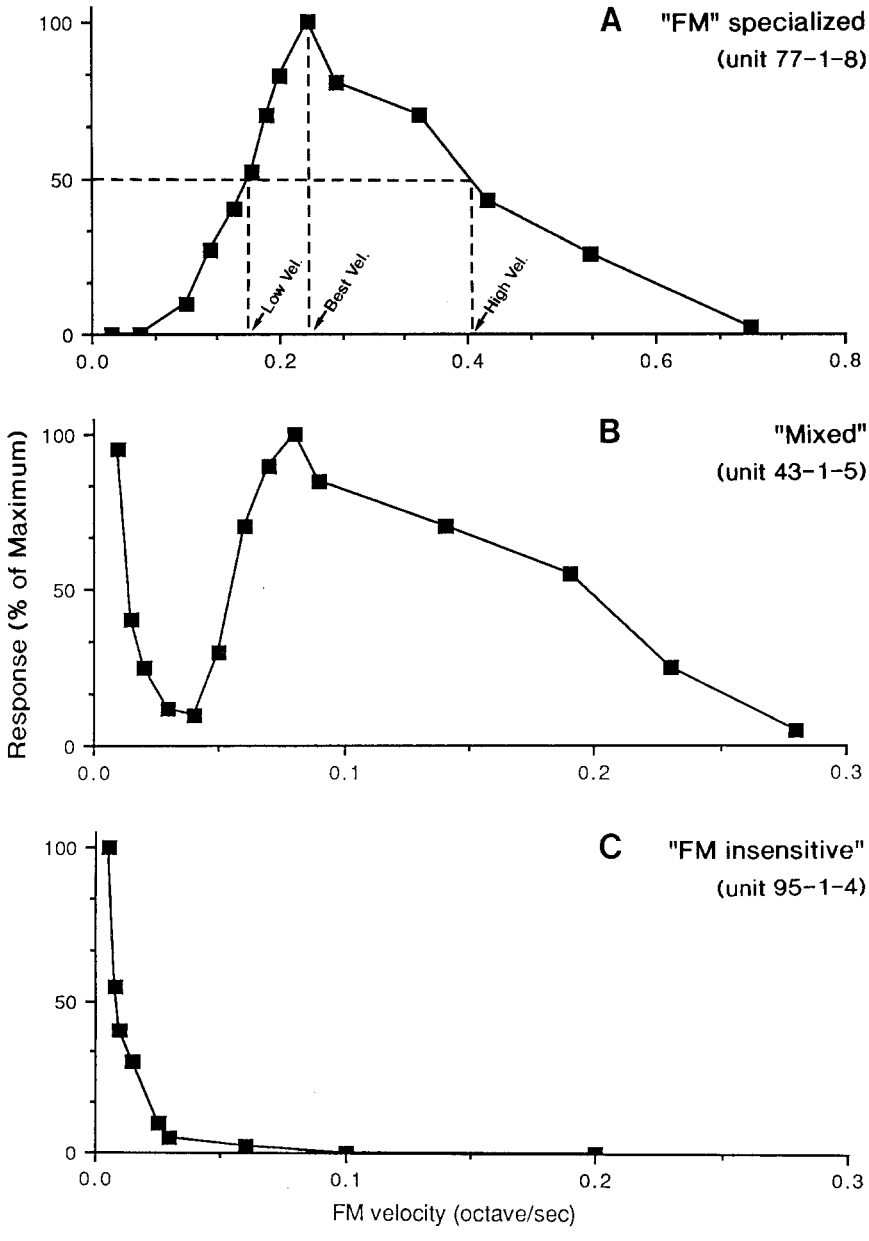


**Fig. 2.** Response functions of the three types of cell responses found at the IC. **A** FM specialized cell; same data as in Fig. 1 but plotting response function as relative response magnitude against FM velocity. The tuning factor, based on intercepts at half-maximum response level, was expressed as Q value [best velocity/(high velocity − low velocity)]. **B** Response function of a mixed cell which shows an FM component above 0.04 octave/s and a pure tone component below that. The FM and pure tone components are typically separated by a dip in the response function. Mixed cells often show a more or less symmetrical FM component corresponding to response to both rise and fall phases of the FM ramp. **C** Response function of an FM insensitive cell showing only the pure tone component

FM cells in relation to a number of response measures: FM velocity, FM range, FM intensity and their respective Q values.

It is well known that IC neurons can be grouped into several morphological categories (Faye-Lund and Osen 1985; Morest and Oliver 1984; Ryugo 1976). However, little is known about how these morphological groups correspond to group differences in response to sounds. In the second part of this study, we described the morphology of a number of physiologically identified IC cells following intracellular filling with horseradish peroxidase (HRP).

This work has been presented in abstract form (Poon et al. 1990a).

## Materials and methods

### *Experimental preparation*

Our methods have been described in detail in a previous report (Poon et al. 1991). Briefly, adult rats (290–310 g) were anaesthetized with sodium pentobarbital (50 mg/kg i.p., supplemented at 15 mg/kg i.p. when necessary during the experiment). A screw was cemented on the skull for holding the animal in a stereotaxic frame. The IC was exposed by a craniotomy and aspiration of the occipital lobe while cerebrospinal fluid was drained at the foramen magnum. Agar was applied topically to control brain pulsation.

### *Acoustic stimulation*

The experiment was performed in an acoustic chamber. Three types of test signals were used: (1) clicks, produced by a rectangular pulse (0.01 ms in width); (2) continuous pure tones, produced by a signal generator (Tektronix FG501); and (3) FM tones, triangularly modulated at 2/s, symmetrical in their rising and falling phases, derived from cascading two function generators (Tektronix FG501A connected through an attenuator to the VCF input of a Tektronix FG501; Fig. 1A). FM range was varied in dB steps (from 0 to 110 dB) and FM velocity in continuous steps corresponding to 0.2–500 Hz of the triangular modulation rate. The free field speaker (Pioneer SEL15) was placed 60 cm in front of the animal, corresponding to zero degree in azimuth and elevation. The overall frequency response of the audio system was within ± 10 dB from 0.5 to 20 kHz (calibration curve given in Poon et al. 1991).

### *Electrophysiological recording*

Unit activity was recorded extracellularly with glass micropipettes (5% NaCl, 10–30 MΩ) advanced vertically through the IC with a stepping microdrive (Narishige). Spike activity was amplified (HV Electrometer 400 B), conditioned to TTL pulses, and sampled by a microcomputer (IBM/AT) to generate PSTHs (peristimulus time histograms).

After an auditory unit had been identified, its MT (minimum threshold) and BF (best frequency) to continuous tones were determined audio-visually. According to our previous definition, mixed but not FM specialized cells responded in a sustained fashion to pure tones below the maximal intensity delivered by the acoustic system. Subsequently, the two types of FM cells were characterized by their response to changes in FM velocity and FM range. The best FM range and velocity of a unit were first determined manually, then the FM range was fixed at that value appearing best for exciting the cell while stimulus level was held at 5–15 dB suprathreshold to its MT. Subsequently, FM velocity was varied systematically around this best value with the tone sweep centred at the cell's BF. After determining a best FM velocity, i.e. that at which the cell's response was maximal, the FM range was varied with the FM velocity maintained at its best setting. The procedure involved holding one parameter constant while systematically changing the others.

In the case of an FM specialized cell, its BF was estimated from the values of other units in the same track, i.e. according to the dorso-ventral tonotopic organization (Clopton et al. 1973; Coleman et al. 1982; Huang and Fex 1986; Ryan et al. 1988).

### *Data analysis*

For most neurons, the data obtained included the following.

*Best FM velocity.* When a cell showed tuning to FM velocity in the form of a non-monotonic (or 'bell-shaped') function, the best FM velocity refers to that producing the cell's maximal response. For cells showing monotonic response functions to FM velocity with a gradual rise to a plateau, the maximal response was taken as the response level at the maximal FM velocity tested. FM velocity, in this study, was expressed as the change in octave/s, instead of absolute Hz/s, to allow comparison between units based on the way frequency is represented logarithmically along the basilar membrane (Kiang et al. 1976; Liberman 1984).

*Best FM range.* This corresponds to the FM range (or depth of modulation, again expressed in octaves) producing the cell's maximal response (sum of both rising and falling responses in the PSTH). For units showing a monotonic response function to FM range, the maximal response was taken as the response at the greatest FM range tested.

*Bandwidth of tuning to FM velocity.* To compare the sharpness of tuning to FM velocity between different FM cells showing non-monotonic response functions, a Q value was computed. This value was equal to the best FM velocity divided by the bandwidth of FM velocity at the half-maximal response level (Fig. 2A).

*Bandwidth of tuning of FM range.* Similar to the estimation of tuning to FM velocity, a Q value for the FM range was also computed for those cells showing a non-monotonic response function to the variation in FM range.

*Threshold FM intensity.* This refers to the minimal intensity level of the FM signal needed to evoke a threshold response of the cell as determined audio-visually.

*Response latency to the click.* This is the interval between the onset of the fixed intensity click (after accounting for free field transmission delay) and the first spike latency in the PSTH.

*Response symmetry.* We followed Britt and Starr's (1976) original method of calculating the symmetry factor (s). Briefly, at the best FM velocity of a given cell, the difference in magnitude of response (area under the curve in the PSTH) to the rising and falling phases of the tone sweep was calculated and then divided by the sum of the two responses. The result, disregarding the sign, yielded the s value. Cells with s less than 0.8 were classified, according to the convention, as 'bi-directional'; the other cells were classified as 'uni-directional'.

### *Histological verification of the recording sites of single units*

For each electrode penetration in which cells were recorded, two marks, one at the beginning and another at the end of the track, were made by electrophoretic injection of pontamine blue (6 μA for 10 min). At the end of experiment, the rat was killed with an overdose of barbiturate, the brain perfused (10% formalin in saline)

and sectioned at 60 μm on a freezing microtome in the transverse plane. The electrode tracks were reconstructed at a magnification of 17× from the serial sections stained in cresyl violet. Sites of units recorded in the IC were estimated on the basis of recording depths measured from the collicular surface after adjustment for shrinkage due to fixation. The locations of each type of FM cells, for the total population of animals, were displayed in serial brain sections at 240 μm in thickness (details of reconstruction method described in Xu et al. 1990).

### *Intracellular study with HRP*

*Dye injection.* Bevelled micropipettes of tip impedance 40–80 MΩ (Sutter Instrument, BV10), filled with a 6% HRP solution (Sigma type VI, in 0.5 M KCl and 0.04 M TRIS buffer, pH 8.6) were used. FM cells, after being characterized extracellularly, were impaled by advancing the microelectrode in small steps (3 μm). The criteria of a successful penetration included an abrupt negative shift in membrane potential exceeding 40 mV which could be maintained during the dye injection, and a reverse positive shift on the subsequent withdrawal of the micropipette. Cells were stained with HRP by delivering positive current (10 nA, 200 ms on, 50 ms off) for 3 min into the impaled neurons. No more than three neurons of the same FM type were labelled in each animal.

*Histological procedures.* Thirty minutes after the last HRP injection, the deeply anaesthetized rat was perfused with fixatives (1% paraformaldehyde, 2% glutaraldehyde) in neutral phosphate buffer. The brain was removed from the skull, and the block containing the IC was sectioned (60 μm) on a freezing microtome in the transverse plane. After processing with HRP histochemistry (DAB method, Adams 1977; Bishop and King 1982), the sections were mounted on glass slides and counterstained with neutral red (1%) before profiles of the labelled cells were traced with camera lucida in the serial sections at 400× magnification. The number of dendritic spines of a cell was counted from 10 randomly selected segments at a magnification of 1000×.

## Results

A total of 117 rats were used in this study, including 81 from the physiology experiment, and 36 from the HRP injection experiment.

### *Physiological data*

A total of 650 click-driven FM units were found in 105 electrode penetrations through the IC. The proportions of mixed and FM specialized cells were 56% and 44% respectively (corresponding to 365 and 285 units; Table 1). The response latency to clicks ranged from 6 to 20 ms and was inversely related to BF (consistent with the classical study in the auditory nerve using click as a stimulus; Kiang 1975).

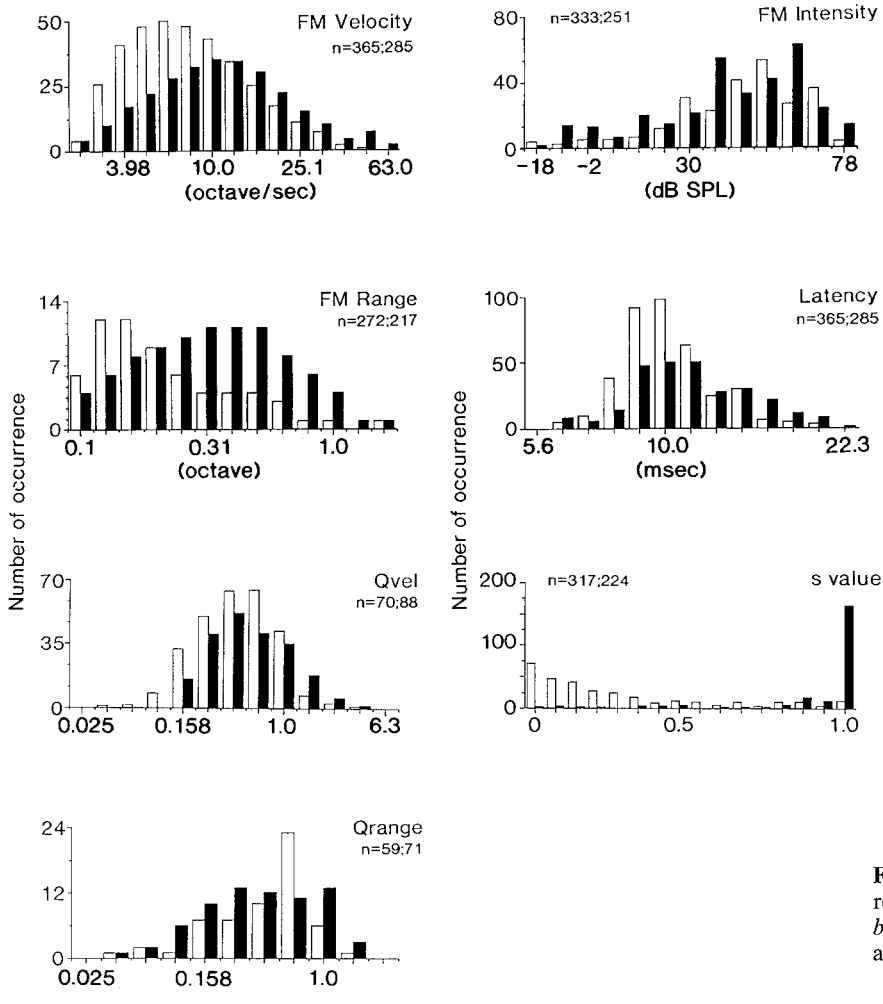


**Fig. 3.** Distribution of characteristics of the seven response measures. *White bars,* mixed cells; *black bars,* FM specialized cells; *n,* numbers of mixed and FM specialized cells in each panel

**Table 2.** Statistical comparison of response characteristics between FM specialized and mixed cells

| Response measure | FM specialized cells | Mixed cells | $p$ (two-tailed) |
|---|---|---|---|
| Best FM velocity (octave/s) | 49.5 ± 6.2 (285) | 18.9 ± 1.6 (365) | < 0.001 |
| $Q_{FM\ velocity}$ | 1.12 ± 0.06 (217) | 0.84 ± 0.04 (272) | < 0.05 |
| Best FM range (octave) | 0.90 ± 0.12 (88) | 0.56 ± 0.06 (70) | < 0.05 |
| $Q_{FM\ range}$ | 0.97 ± 0.09 (71) | 0.63 ± 0.06 (59) | < 0.005 |
| Threshold FM intensity (dB SPL) | 33.33 ± 1.3 (251) | 23.33 ± 1.3 (333) | < 0.001 |
| Response latency to click (ms) | 9.07 ± 0.47 (285) | 7.08 ± 0.30 (365) | < 0.001 |
| Response symmetry (s factor) | 0.889 ± 0.014 (224) | 0.291 ± 0.016 (317) | < 0.001 |

Mean and SEM values of the raw data before logarithmic transformation. The number of cells is given in parentheses (note the numbers are not equal as response measures were not completely characterized for each cell). The $P$ values apply for both raw and transformed data

**Table 3.** Statistical comparison of response characteristics with the alternative classification of FM cells according to response symmetry (s value), showing poorer segregation

| Response measure | Uni-directional cells ($s \geq 0.8$) | Bi-directional cells ($s < 0.8$) | $P$ (two-tailed) |
|---|---|---|---|
| Best FM velocity (octave/s) | 42.1 ± 6.6 (247) | 20.6 ± 2 (339) | < 0.001 |
| $Q_{FM\ velocity}$ | 1.08 ± 0.07 (193) | 0.98 ± 0.05 (286) | > 0.05 |
| Best FM range (octave) | 0.76 ± 0.11 (68) | 0.67 ± 0.07 (91) | > 0.05 |
| $Q_{FM\ range}$ | 1.04 ± 0.212 (53) | 0.67 ± 0.06 (78) | < 0.01 |
| Threshold FM intensity (dB SPL) | 33.4 ± 1.3 (249) | 23.1 ± 1.3 (343) | < 0.001 |
| Response latency to click (ms) | 8.87 ± 0.47 (247) | 7.15 ± 0.32 (339) | < 0.01 |

Same notations as in Table 2

Due to the difficulty in maintaining sufficiently stable recordings, about 75% of the cells were characterized with respect to only five of the seven measures described in Materials and methods; generally the best FM intensity and its Q values were not determined. Complete sets of data were obtained for the remaining 25% of cells. For each response measure, the distribution of values was highly skewed (similar to the data reported by Poon et al. 1991). In order to apply parametric statistics based on a normal distribution, the original data were logarithmically transformed (Fig. 3).

Striking differences ($P < 0.05$, Student's $t$-test) in sensitivity were observed for all response measures examined between the two groups of FM cells (Table 2). The significance of differences in best FM velocity, its Q value, threshold FM intensity and response latency was high ($P < 0.001$) when the sample size was also large. In addition, the difference in s value between the FM specialized and mixed cells was highly significant ($P < 0.001$; Table 2).

In contrast, the difference in response characteristics between the bi-directional and uni-directional cells was less significant (Table 3).

FM specialized cells preferred larger FM velocities, larger FM ranges, and were more narrowly tuned (i.e. had larger Q values) to both FM velocity and FM range than were the mixed cells. FM specialized cells also responded to FM tones at higher intensity levels, and were activated by click at longer latencies. Finally, nearly 80% of the FM specialized cells were uni-directional.

With the exception of response symmetry, hardly any correlation existed between the response characteristics. The highest correlation ($r = 0.31$) was found between the FM velocity and FM range.

### *Regional distribution of the FM cells in the IC*

Recording sites of 488 FM cells (from a total of 650) were successfully reconstructed. These cells were located at

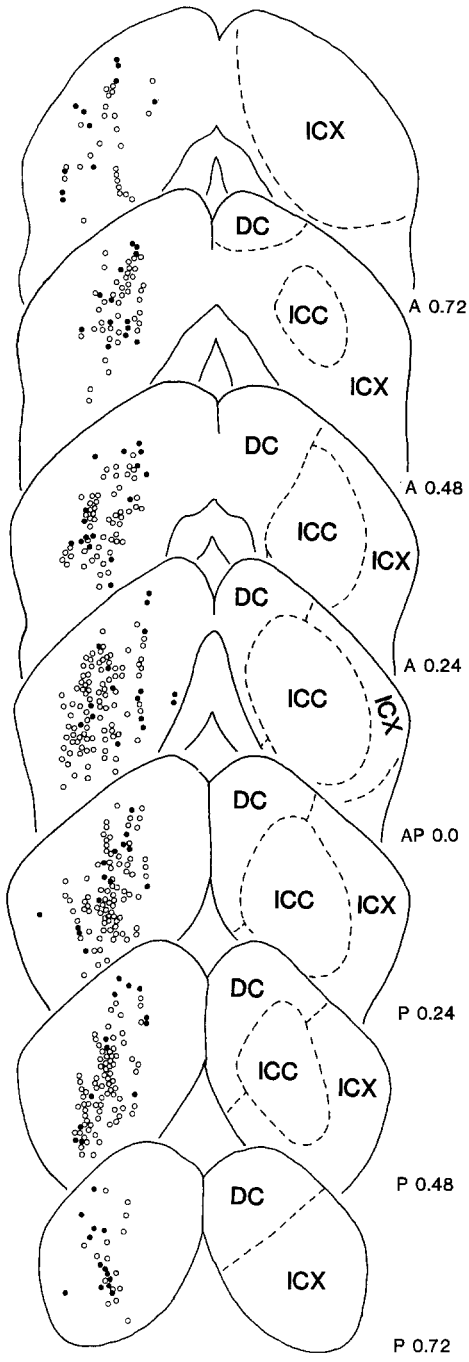


**Fig. 4.** Regional distribution of the 488 FM cells in the IC seen in serial sections in the coronal plane. The nuclear subdivisions of IC were retraced on the right side with reference to Paxinos and Watson (1982) and Faye-Lund and Osen (1985). Locations of individual units are represented by *empty circles* for mixed cells and *filled circles* for FM specialized cells. **A, P,** distance (millimetres) anterior or posterior to the interaural plane; *DC,* dorsal cortex; *ICC,* central nucleus; *ICX,* external nucleus

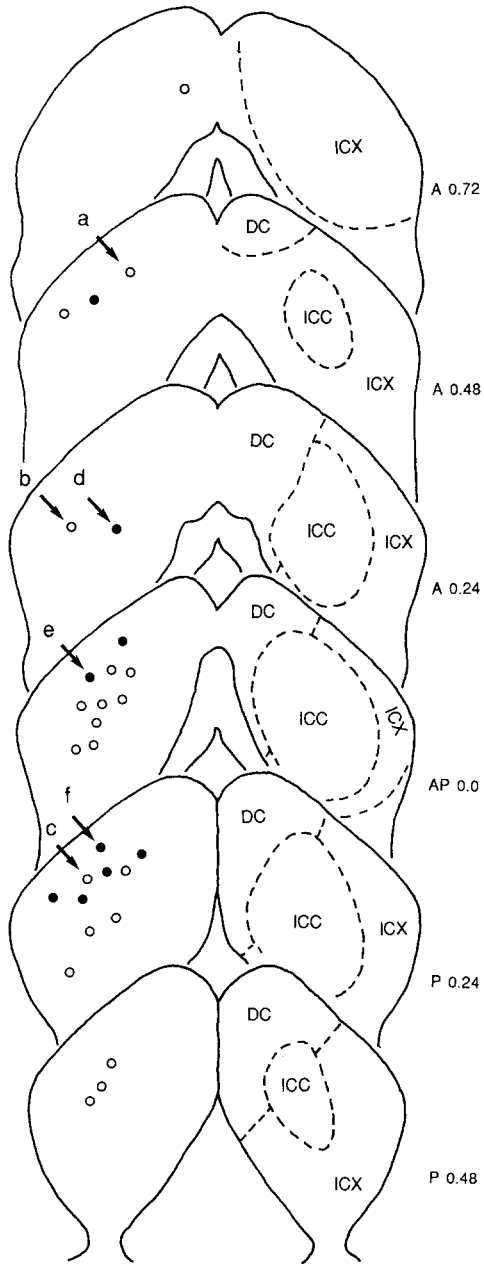


**Fig. 5.** Locations of 29 HRP filled FM cells in the IC matched to 6 coronal sections. Nuclear subdivisions and notations are the same as in Fig. 4. The *arrows* indicated the cells whose profiles are traced in Fig. 6

depths between 0.3 and 3.0 mm below almost the entire surface of the exposed IC. Nearly 95% of cells were found inside the two major gross subdivisions of IC (Aitkin 1986): the central (ICC) and external (ICX) nuclei (Fig. 4). Both types of FM cells were present in ICC as well as ICX (32.6% of FM specialized cells in ICC, 56.5% in ICX; 48.7% of mixed cells in ICC, 47.3% in ICX). Only a small proportion of FM cells were located in dorsal cortex (DC, or dorsomedial nucleus; Rockel and Jones 1973). There was no clear segregation of the FM cell types according to the three gross anatomical subdivisions of the IC, or any local clustering of a given type of FM cells, but more FM specialized cells were found in the ICX. There was no apparent difference in response properties between FM cells found in the ICC and ICX.

We also examined the regional distribution of 132 FM insensitive cells (from a total of 194) found in the same recording tracks (results not shown here). These cells were found only in the ICC and ICX, with higher incidence in the ICC.

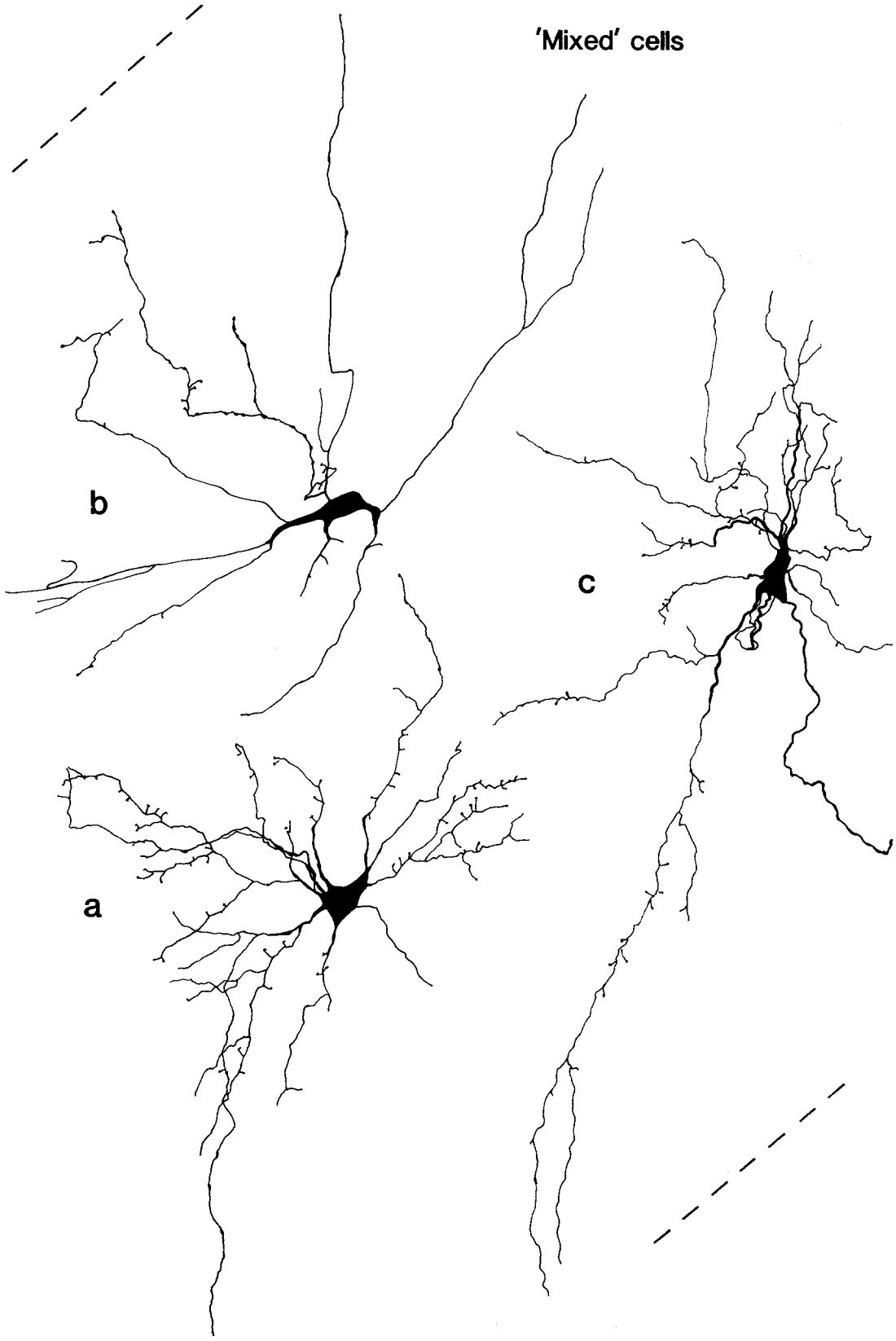


**Fig. 6A–F.** Camera lucida drawings of some mixed cells (**A–C:** units 149–2–1, 151–1–1, 144–1–1), and FM specialized cells (**D–F:** units 145–1–1, 155–1–1, 131–4–1) the locations of which in the IC are indicated by arrows in Fig. 5. For comparison, the cells were traced under the same magnification, and placed in the same co-ordinates (as marked by the dorsal-lateral scale). Note that FM specialized cells had more extensive dendritic arborizations and more dendritic appendages. The dendrites appeared to orient across the isofrequency laminae, which, for the purpose of comparison, were estimated to run parallel to the dashed lines

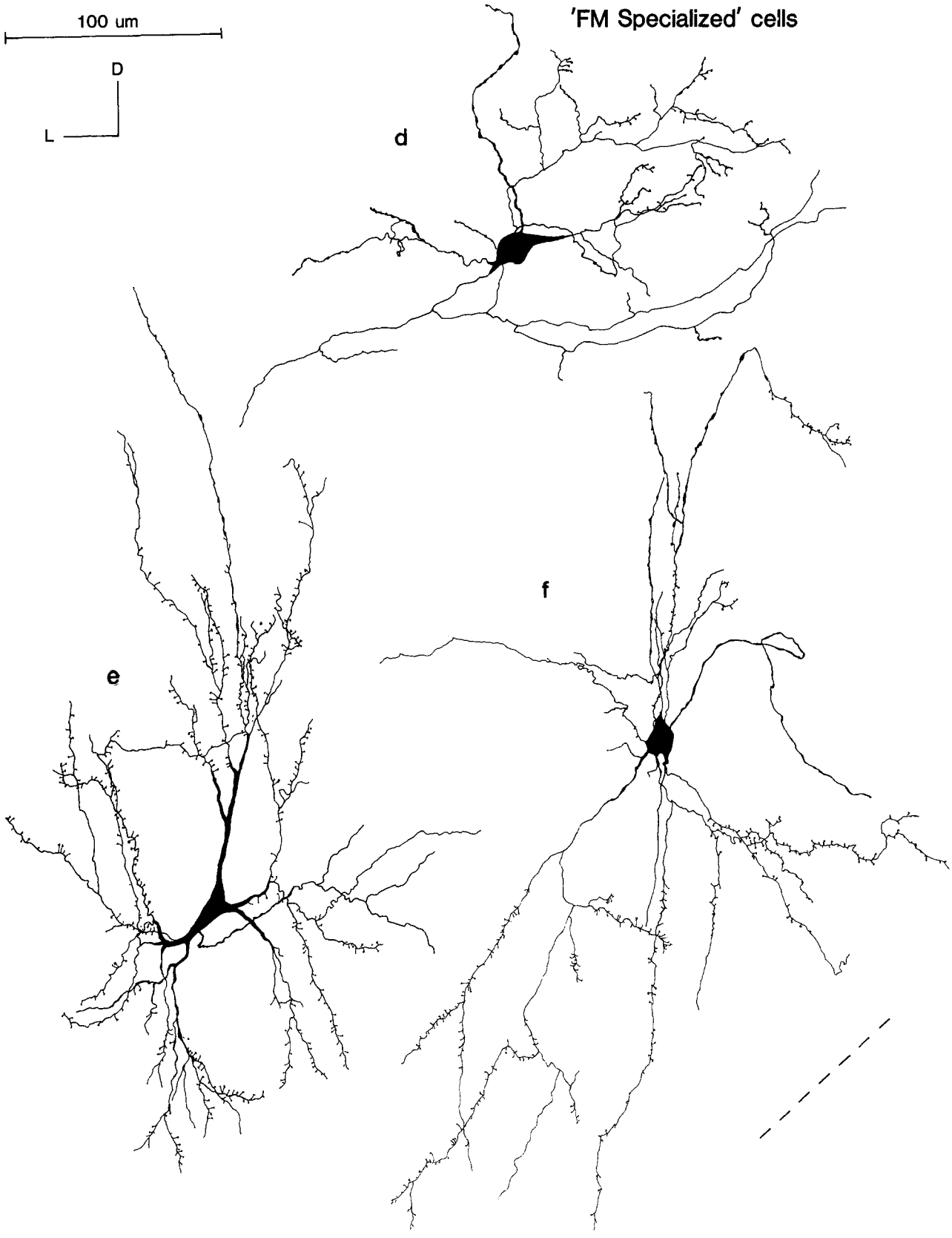


Fig. 6

*HRP histological data*

Twenty-nine FM cells were filled. Of these, 20 belonged to the mixed and 9 to the FM specialized category. They were found in ICC and ICX, and their distribution in the IC (Fig. 5) was consistent with those of a larger population of IC cells (Fig. 4). More FM specialized cells were found in the ICX.

All FM cells were multipolar in appearance with relatively broad dendritic fields (150–450 μm). These cells corresponded to the multipolar cells described in the literature (Rockel and Jones 1973; Ryugo 1976) or the stellate cells (Oliver and Morest 1984). Consistent with previous reports, the dendrites of these cells were oriented in a direction perpendicular to the isofrequency laminae. Striking differences in cytomorphology were found between the two groups of FM cells. Specifically, the FM specialized cells had more dendritic spines, larger dendritic fields, and more dendritic branchings (Fig. 6). A quantitative comparison of the cytomorphological features is shown in Table 4.

Axons of all the labelled cells could also be differentiated. They did not appear to be interneurons. Most of their axons left the IC and entered the brachium of the same side, while others were found to terminate locally in the IC. There was no apparent relationship between the destination of the axons and the type of FM cells.

For 20 labelled FM cells, the best FM ranges were also determined. The CFs of these cells ranged from 2.1–45.2 kHz (mean 12.2, SD 12.8 kHz). A somewhat linear relationship was found between the dendritic extent and the best FM ranges in these cells (Fig. 7). There was no apparent relationship between the pattern of dendritic arborization and the response symmetry.

We also examined the cytomorphology of a very limited number of FM insensitive cells. They had typically small amplitude extracellular potentials, and were difficult to impale with our micropipettes. When they were successfully injected with HRP, only a few of them (3 of 13) could be stained. They were found in the ICC and were bipolar neurons of small size (dendritic extent $< 150$ μm), corresponding to the main principal cell type in the ICC (Rockel and Jones 1973; Ryugo 1976).

**Table 4.** Statistical comparison of cytomorphological features between FM specialized and mixed cells

| Cytomorphological feature | Mixed ($n=20$) | FM specialized ($n=9$) | p |
|---|---|---|---|
| Dendritic extent across isofrequency laminae (μm) | 165 ±65 | 227 ±67 | <0.05 |
| Number of primary dendrites | 3.3± 0.6 | 4.0± 0.8 | >0.05 |
| Number of secondary dendrites | 6.5± 0.9 | 12.7± 1.9 | <0.001 |
| Dendritic spine density (number/mm) | 45 ±34 | 146 ±59 | <0.001 |

Data are expressed as mean ± SD. Differences are significant except for primary branchings, where the number is small

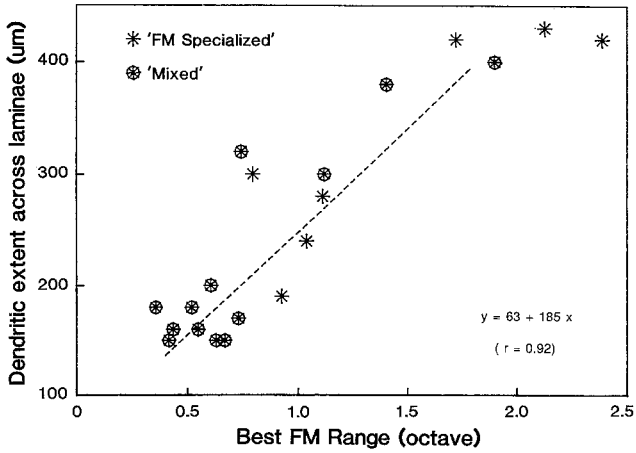


**Fig. 7.** The relationship between physiologically characterized best FM range and the histologically determined dendritic extent in 20 FM cells. *Dashed line* represents the linear regression of these points

## Discussion

The primary contrast between the FM specialized and mixed cells lies in their differential response to pure tone stimulation. Although the pure tone response we described was obtained with continuous stimulus, instead of tone pips as used by other investigators, our results appeared comparable to the findings of previous studies (e.g. we obtained MTs below 0 dB sound pressure level, SPL). Furthermore, the difference in pure tone threshold between the two types of FM cells was large enough to ensure a robust classification even with adaptation. The secondary contrast was that FM specialized cells are more narrowly tuned to all parameters of the FM stimuli. The 'FM specialized' cells may thus represent a group of cells more specialized for coding FM signals. Suga (1965a) postulated the existence of recurrent and lateral inhibition on the FM cells of the bat IC. Models for FM cells in the IC based on combined excitatory and inhibitory inputs have also been proposed (Erulkar et al. 1968). A similar model has also been applied to FM coding in the cochlear nucleus (Fernand 1971). If FM coding indeed takes place at the IC, as suggested by the emergence of a large proportion of FM cells, some specialized arrangement of synaptic inputs from adjacent isofrequency laminae may generate the highly characteristic response to FM sweeps including the asymmetrical response. FM cells at the IC had broad dendritic fields and the dendrites appeared to lie against an orderly array of afferent terminals. Mixed cells, by virtue of their sensitivity to pure tones, are perhaps only partially furnished with such inhibitory circuits and are therefore less specialized in detecting FM features.

The principal finding of the intracellular injection experiments was that the physiological differences found between FM cells were correlated with cytomorphological differences. Because of the wide overlap in distribution between the FM specialized and mixed cells, the statistical differences found between their response characteristics may be considered at the population, rather than individual, level. All the morphological differences between the IC neurons have been previously reported in the literature (Faye-Lund and Osen 1985; Morest and Oliver 1984; Ryugo 1976). The difference in

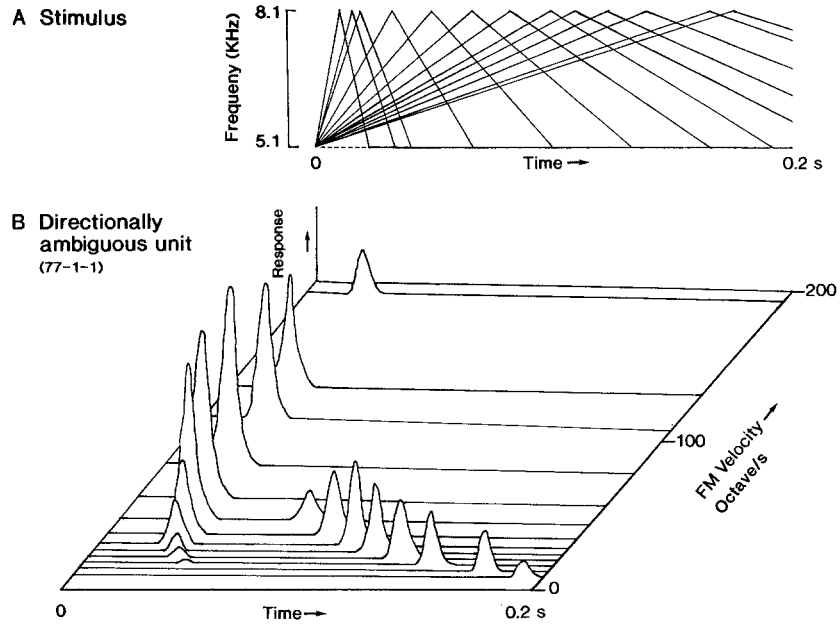


**Fig. 8A, B.** Example of a directionally ambiguous unit. **A** The modulating signals of the FM stimulus; **B** the corresponding PSTHs. At high FM velocities, the bi-directional response became uni-directional

size of dendritic fields is of particular interest. In addition to a possible relationship between dendritic extent and the number of dendritic branchings, large dendritic fields of FM cells were associated with large FM ranges. As the dendritic fields of the multipolar neurons in the IC are roughly aligned perpendicular to the isofrequency laminae (Oliver et al. 1991), it is tempting to speculate that synaptic inputs to the FM cells can be derived from afferent fibers originating from more than one isofrequency laminae. The implication of this is that FM coding could take place at the IC primarily by the convergence of adjacent inputs.

That fundamental differences exist between the two types of FM cells was further reflected in their differences in response latency to sound. With the click stimuli, FM specialized cells were activated at a longer latency (on average 2 ms longer) than the mixed cells. The same conclusion could be drawn with respect to the response latency to FM tones. Using a linear regression method (Whitfield and Evans 1965) we analysed the PSTHs obtained at different FM velocities, and found almost the same difference in central transmission time between the two types of FM cells. Such difference in latency is not due to the difference in BF between the two types of cells since their BFs are similar in distribution. The difference in latency is probably due to: (a) the addition of one or more interneurons in the pathway, and/or (b) a difference in the integrating properties of the cells. Future experiments with intracellular recordings from the IC will be one way to differentiate between these two possibilities.

Classifying FM cells according to response symmetry is subject to the following criticisms: (a) for the same cell, varying FM velocity alone could lead to different s values (Fig. 8). In some cases, the response symmetry was altered when the same FM ramp was inverted, as would be expected from adaptation to successive stimuli. (b) The distinction between the uni-directional and bi-directional cells was not clear cut (Fig. 9). However, there existed a strong statistical linkage ($P < 0.001$) between the mixed and bi-directional cells and between the FM specialized

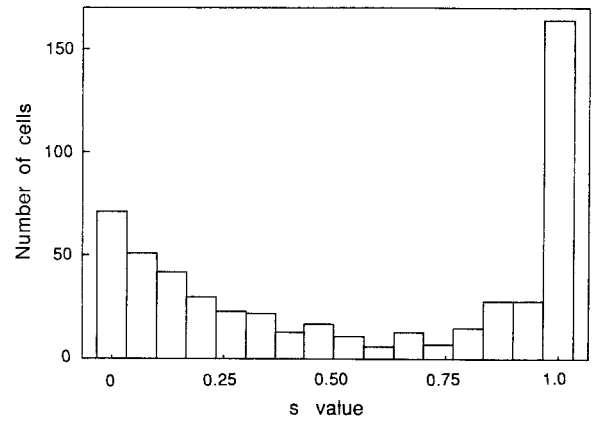


**Fig. 9.** Distribution of response symmetry factors (s) in 541 FM cells. Though the histogram is clearly bi-modal, the boundary is ill defined, as there is a smooth transition in s values between the two extremes

and uni-directional cells. Consequently, it was not surprising to find that the segregation of FM cells, based on response symmetry, yielded similar but less significant results.

Most cells with similar BFs were found at similar depths below the surface. The gross tonotopic laminations appeared continuous throughout the ICC and ICX, similar to that previously reported for metabolic markers (Huang and Fex 1986; Coleman et al. 1982; Ryan et al. 1988), as well as microelectrode mapping (Clopton and Winfield 1973; Stiebler and Ehret 1985). Such continuity of response characteristics spreading through ICC and ICX may explain the somewhat unexpected finding that the FM cells found in these two subdivisions appeared physiologically and morphologically similar.

The significance of the presence of more dendritic appendages or dendritic spines on the FM specialized cells remains unclear. Our observation that spiny cells have more higher order branches was consistent with other studies (Morest and Oliver 1984; Oliver et al. 1991). The abundance of dendritic appendages could

reflect a more complex synaptic organization associated with their more narrowly tuned properties to FM stimuli, as well as a possible relationship to neuronal plasticity at the IC to neonatal acoustic experience with complex sounds (Poon et al. 1990b).

A systematic study of the FM sensitivity using a similar approach at different levels, e.g. at the cochlear nucleus and the auditory cortex, has not yet been undertaken. The fact that FM sensitivity is specific to the level of the IC, while FM specialized cells are not found at lower levels (such as the cochlear nucleus; Sinex and Geisler 1981), suggests that IC may be an important centre for encoding FM sounds that are often present in speech signals (Kay 1982). The full implications of our findings on cytomorphological correlates and extracellular responses for the understanding of FM coding in the IC may only become clear with further studies using intracellular recordings from these cells.

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