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Structure and Function of Electrosensory Neurons in the Torus Semicircularis of *Eigenmannia*: Morphological Correlates of Phase and Amplitude Sensitivity[1]

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Abstract

Structure-function relations in the electrosensory system of *Eigenmannia* were examined by labeling physiologically characterized neurons of the dorsal torus semicircularis. The sensitivity of cells to modulations in amplitude (AMs) and modulations in differential phase was determined. Approximately half of the 48 cell types defined by Golgi studies (Carr, C. E., and L. Maler (1985) J. Comp. Neurol. *235:* 207–240) were identified in this manner. The majority of the neurons located in laminae (8a, b, c, and d, and 9 exhibited sensitivity to differential phase. In laminae 5 and 7, however, in addition to neurons which were sensitive to differential phase, many cells were found that were purely AM sensitive. Differential phase sensitivity originates in the small cells of lamina 6 (Heiligenberg, W., and G. Rose (1985) J. Neurosci. *5:* 515–531), the exclusive termination site of phase-coding afferents from the electrosensory lateral line lobe. Cells that had dendritic extensions into the neuropil of lamina 6 exhibited sensitivity to differential phase, whereas neurons lacking dendrites in this lamina were only excited by AMs.

These findings support the notion of a relationship between the morphology and laminar position of a neuron and its function.

Sensory information is commonly processed in laminated structures of the CNS. This form of structural organization may, therefore, be relevant to perceptual mechanisms. Although single-unit studies hint at a functional differentiation of laminae, it is still unclear how strongly the morphological specializations of individual neurons are linked to their functional properties (Gilbert, 1983).

Recently, various investigators (Heiligenberg and Dye, 1982; Saunders and Bastian, 1984; Heiligenberg and Rose, 1985; C. E. Carr, B. Taylor, and L. Maler, submitted for publication) have utilized intracellular recording and iontophoretic techniques to investigate structure-function relations in the electrosensory system of weakly electric fish. Most studies have used *Eigenmannia*, an electric fish from South America. This species can be bred in the laboratory, thus ensuring a large supply of experimental animals. Furthermore, a great deal is known about the stimulus parameters which are important in controlling electrosensory behaviors and about the response properties of electrosensory neurons in the CNS.

One behavior, the jamming avoidance response (JAR), has been particularly well studied (Bullock et al., 1972; for review, see Heiligenberg, 1983). In this behavior, the fish shifts its electric organ discharge (EOD) frequency away from that of a neighbor. When the two EODs are similar in frequency, their addition forms a nearly sinusoidal signal which "beats" at the difference frequency, *Df*, of the interfering EODs. This beating signal is characterized by a periodic modulation of the amplitude (AM) and of the phase (the timing of the positive zero-crossings of the beating signal, relative to the zero-crossings of either EOD alone). The degree of contamination of the fish's EOD signal by the neighbor's EOD signal will vary over the body surface; the region being most heavily contaminated is a function of the relative orientation of the two fish. Although the animal can determine the magnitude of *Df* from the rate of the AM alone, discrimination of the sign of *Df* (i.e., whether the neighbor's EOD frequency is higher or lower than the animal's own EOD frequency) requires that it monitor AMs within a particular area of the body and differential phase between this area and other areas that are less strongly contaminated by the interfering signal. Specifically, with the neighbor's EOD having a lower frequency, the area of the animal's body surface with the largest signal modulation will experience a rise in amplitude during a phase lead and a fall in amplitude during a phase lag of this signal in reference to the signal at some other part of the body surface. The reverse association holds if the neighbor's EOD frequency is higher (Heiligenberg, 1980).

In their earlier extracellular work, Bastian and Heiligenberg (1980a, b) and Partridge et al. (1981) provided evidence that differential phase sensitivity first arises in the torus semicircularis of the midbrain. In subsequent intracellular work (Heiligenberg and Rose, 1985) we identified the small cells of lamina 6 as the origin of differential phase sensitivity. This finding is supported by ultrastructural evidence (C. E. Carr, B. Taylor, and L. Maler, submitted for publication).

The present study is a continuation of our labeling of physiologically identified neurons in the electrosensory system. We have identified cell types which are sensitive to AMs, modulations in differential phase, or both, and we are now able to relate such functional properties to the laminar location and dendritic organization of these neurons.

Materials and Methods

Our experimental procedures followed those used in earlier intracellular studies (Heiligenberg and Dye, 1982; Heiligenberg and Rose, 1985). Animals of the genus *Eigenmannia*, ranging in length from 8 to 18 cm, were immobilized by intramuscular injection of 1 to 2% Flaxedil, gently suspended under the water surface, and respirated by a constant flow of water through their mouths. A small Plexiglas holder was glued to a parietal bone to mechanically stabilize the head for intracellular recording. A small hole, less than 1 mm in diameter, was drilled through the thin bone covering the optic

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tectum, and a silver ground wire was placed at the edge of this hole. Electrodes were pulled on a Brown-Flaming micropipette puller and their tips were filled with a 10% solution of Lucifer Yellow in 0.1 M LiCl. These electrodes had a resistance of 200 to 400 megohms.

Neurons were penetrated by applying a brief positive current pulse. Responses of a neuron to various stimulus regimens were tape-recorded, and the neuron was then filled by iontophoretic injection with a DC-offset sine wave current, at a frequency of 3 Hz and a peak-to-peak amplitude of 2 nA of negative polarity. Cells were injected for 1 to 10 min.

Since the animal's EODs are silenced by injection of curare-like drugs, its electroreceptors can be driven exclusively by artificial electric stimuli. Much as in previous experiments, we substituted for the animal's EOD a sine wave stimulus (S_1) applied between an internal electrode ("gut electrode") and an external electrode (at the tail), so that current, much as in the case of the natural EOD, penetrated the skin fairly perpendicularly and with the same polarity. The stimulus voltage was adjusted to that of the animal's own EOD prior to curarization. A second sine wave stimulus (S_2) was presented either through the same pair of electrodes as was S_1 ("identical" stimulus geometry) or through any two of a circular array of electrodes surrounding the fish ("differential" geometry).

The responses of individual neurons were analyzed by constructing histograms wherein spike activity was plotted as a function of the AM (beat) cycle. Amplitude modulation cycles of opposite signs of *Df* are characterized by opposite modulations in differential phase. Phase-sensitive neurons will, therefore, fire maximally at different periods of the beat cycle for positive and negative *Df* situations. Conversely, for purely AM-sensitive units, beat cycle histograms will be nearly identical for the two signs of *Df*. Sensitivity to AM was calculated from spike histograms obtained under identical stimulus geometry (i.e., modulations of stimulus amplitude without modulations of differential phase). The envelope of a histogram was Fourier analyzed. The magnitude of the coefficient at the fundamental frequency of the beat cycle was used as a measure of the unit's AM sensitivity.

Sensitivity to differential phase was calculated using the histograms obtained under differential stimulus geometry. For a given S_2 orientation, the normalized histograms for the two signs of *Df* were subtracted from one another, thereby minimizing the amplitude-dependent component of the response and enhancing the phase-dependent component. The envelope of the resulting histogram was Fourier analyzed. The magnitude of the coefficient at the fundamental frequency of the beat cycle was used as a measure of the unit's sensitivity to differential phase.

Results

Background. The dorsal torus semicircularis consists of 12 laminae. Lamina 6 is the exclusive recipient of spherical cell afferents from the electrosensory lateral line lobe (ELLL) (Fig. 1). The spherical cells in turn receive input from T-type primary afferents. "T" units fire one spike, time-locked to the zero-crossing of the signal, for each stimulus cycle. Two types of pyramidal cells of the ELLL receive amplitude information via P-type primary afferents: basilar pyramidal neurons are excited by an increase in stimulus amplitude, and nonbasilar pyramidal cells are excited by a decrease in stimulus amplitude. Both types provide input to laminae 3, 5, 7, 8c, and 8d of the torus. Lamina 8b is the exclusive toral recipient of a projection from the lobus caudalis (electrosensory cerebellum). Other inputs to the various laminae of the torus are described by Carr et al. (1981) and are not pertinent to this report.

Physiology and anatomy of toral neurons. Golgi studies by Carr and Maler (1985) have identified 48 distinct cell types. By intracellular iontophoresis of Lucifer Yellow, we have labeled approximately half of these types. Prior to labeling a cell, we determined its sensitivity to AMs and modulations in differential phase (i.e., relative timing of signals received by different regions of the body surface). An example of a cell which is driven only by modulations of differential phase is provided in Figure 2. Amplitude modulations had little effect on the activity of this unit (Fig. 2*e*). This neuron, a multipolar cell of lamina 8b, responded most strongly when the S_2 stimulus was presented with transverse orientation. This orientation yields maximal phase modulations between the right and left sides of the body (Heiligenberg and Rose, 1985). The phase modulation functions in Figure 2 reflect, for the transverse orientation of S_2, the timing of the signal on the left side of the fish's body relative to the timing on the right side; advance is up, delay is down. This unit was maximally excited when the signal on the fish's left side was advanced relative to the signal on the right side.

The lamina 8a neuron displayed in Figure 3 is representative of a purely AM-sensitive unit. For each orientation of the S_2 stimulus, the distribution of spikes within the beat cycle is similar for negative and positive values of *Df* and, thus, is independent of differential phase. Most toral neurons were excited by both types of modulation. An example of this type of unit is provided in Figure 4. This large multipolar neuron of lamina 5 responded to a rise in stimulus amplitude (Fig. 4*f*). The pattern of spikes within the beat cycle is most strongly phase dependent for the oblique S_2 orientation in Figure 4*d*, indicating that this neuron compares the timing of the signal on the right, caudal portion of the body surface with the timing on the left, rostral portion.

Cell types that were excited by AMs but were not sensitive to differential phase are presented in Figure 5. Neurons that were sensitive to differential phase are displayed in Figure 6. In general, cells of laminae 8a, 8b, 8c, 8d, and 9 were found to be sensitive to differential phase. In contrast, the more dorsal laminae contain, in addition to phase-sensitive units, neurons which are purely AM sensitive. We have found that this physiological variation can be related to morphological differences between the various cell types of these laminae. Specifically, the analysis of neurons in laminae 5 and 7 suggests that dendritic connections to lamina 6 are necessary for phase sensitivity, which originates in the small cells of lamina 6 (Heiligenberg and Rose, 1985).

Carr and Maler (1985) identified three cell types in lamina 5. All three have been labeled in the present study. Type a (see Fig. 5) is a multipolar cell with a soma of 10 to 15 μm. The dendrites of this cell type are largely limited to lamina 5 but can extend as far as laminae 3 and 4; no dendrites were ever seen encroaching into lamina 6 or the vertical columns of neuropil which penetrate this lamina. The three type-a cells labeled were purely AM sensitive. Type b is a bilaminar cell, having its dendrites in laminae 5 and 7. A great deal of variation exists with regard to the details of this neuron's dendritic arbors. Some type-b cells have ventral dendrites which penetrate deeply into lamina 7, whereas others restrict their ventral dendrites to the vertical neuropil columns. In addition, some bilaminar cells send dendrites into lamina 6. Of the six type-b cells which were labeled, two were phase sensitive. Both of these neurons had dendritic arborizations in the neuropil (containing the giant and small cells) of lamina 6. In addition, two lamina 5 neurons were labeled which were morphologically distinct from the Golgi types described by Carr and Maler (1985). These large, multipolar cells sent dendrites dorsally into lamina 4 and ventrally into lamina 6 (Fig. 6). Both cells were sensitive to modulations in amplitude and differential phase (see Fig. 4). A single type-c neuron was labeled (see Fig. 9). This unit was strongly AM sensitive; its phase sensitivity could not be determined.

The vertical neuropil zones which penetrate lamina 6 contain, in addition to fiber bundles, a very characteristic cell type (Vn, Fig. 5). These vertical neuropil neurons are bipolar and have dendritic arborizations in laminae 5 and 7. The actual position of the soma varies in its proximity to these laminae. The three labeled, vertical neuropil cells only responded to AMs.

From Golgi material, Carr and Maler (1985) identified five types of neurons in lamina 7. With the exception of the "type-d" cell, a smooth multipolar neuron which is believed to be rare, all cell types have been labeled. In addition, a cell type (type f, Fig. 6) was labeled that could not be matched to any of the types described by Carr and Maler (1985). This cell type, having a round soma approximately 10 μm in diameter, is distinguished from other lamina 7 neurons by the numerous dorsal dendrites which penetrate lamina 6. The two type-f neurons that were labeled exhibited moderate sign selectivity (i.e., they responded more vigorously to one sign of *Df* than to the other sign). The physiology of one of these cells is shown in Figure 7. This unit responded best for a negative *Df*. This selectivity was strongest when S_2 was presented in the longitudinal direction (Fig. 7, *top set*

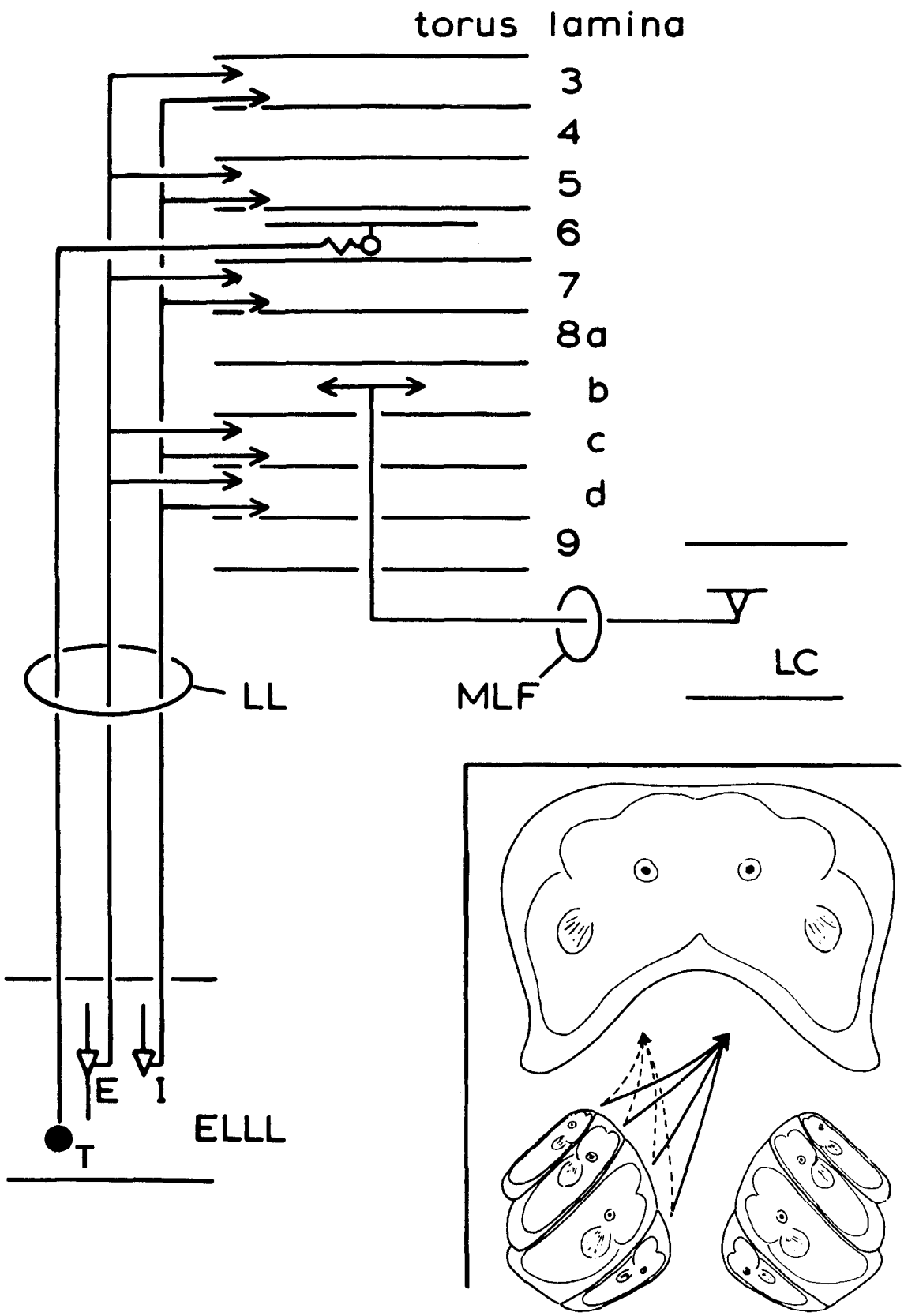

Figure 1. Afferents of the torus semicircularis. The torus receives a predominantly contralateral input, via the lateral lemniscus (*LL*), from the electrosensory lateral line lobe (*ELLL*). Laminae 3, 5, 7, 8c, and 8d receive input from the basilar (*E*) and nonbasilar (*I*) pyramidal cells of the ELLL. "E" units are excited by increases in stimulus amplitude, whereas "I" units respond to decreases in stimulus amplitude. Spherical cells (*T*) of the ELLL terminate solely on the small cells (not shown) and giant cells of lamina 6. "T" units fire one spike per stimulus cycle. Lamina 8b is the exclusive recipient of a projection, via the medial longitudinal fasciculus (*MLF*), from the lobus caudalis (*LC*) of the cerebellum. Neurons from the four maps of the ELLL project onto the torus to form a single map of the body surface (*lower right*; see *symbols* for pectoral fins and eyes).

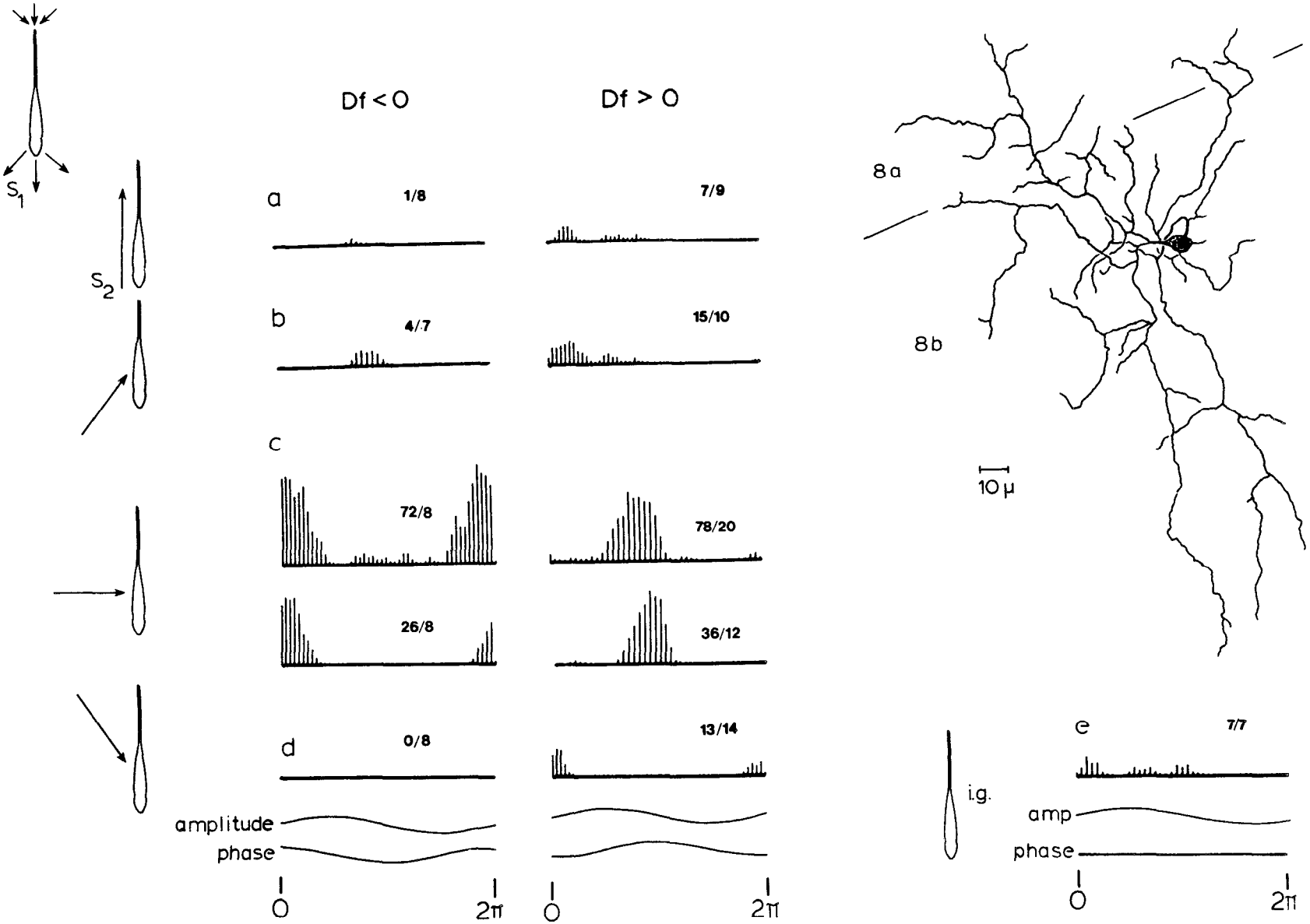

Figure 2. Example of a phase-sensitive unit. The fish's EOD has been replaced by a sinusoidal signal (S_1) of similar frequency, amplitude, and spatial distribution (*inset* in *upper left*; *arrows* indicate positive current flow). A second sinusoidal signal (S_2), which was 2 Hz lower ($Df < 0$) or 2 Hz higher ($Df > 0$) than S_1, was presented either through the same pair of electrodes used for delivering S_1 ("identical geometry" (*i.g.*), in *e*) or through pairs of electrodes straddling the fish in different orientations ("differential geometry," in *a* to *d*). The positive orientation of the S_2 current is indicated by the *arrow* on the *left* in each instance. Data are presented in the form of beat cycle histograms. Each bin represents a period of time equal to three S_1 cycles, 10 msec in this case. The height of each *bar* reflects the probability of a spike occurring in that 10-msec bin. The total number of spikes over the total number of beat cycles is presented for each histogram. Since the beat frequency is 2 Hz, each beat cycle histogram represents 500 msec. These histograms are constructed with reference to the pattern of the amplitude modulation. Therefore, histograms of opposite signs of *Df* have opposite phase modulations. A unit which is sensitive to the timing of the signal in one part of the body relative to the timing of the signal in another region of the body (i.e., differential phase) will, therefore, respond maximally during a different part of the beat cycle for positive *Df* and negative *Df*. For this reason, such a cell is also referred to as "sign sensitive." This multipolar neuron of lamina 8b was only weakly driven by pure AMs (*e*) (note that phase modulations are absent for the identical geometry situation). The unit, however, responded vigorously when the S_2 was presented with transverse geometry (*c*). As can be calculated from the spatial distribution of the S_1 and S_2 currents (Heiligenberg and Rose, 1985), the response maximum occurred during a phase advance of the signal received by the left side of the body relative to the signal received by the right side. The amplitude and phase functions at the *bottom* of the *histograms* indicate AMs and phase modulations at all points in space where the positive vectors of S_1 and S_2 form acute angles (phase advance is up). For all points where the positive vectors of S_1 and S_2 form obtuse angles, these functions must be shifted by half of a beat cycle. A camera lucida drawing of this cell is shown on the *upper right*.

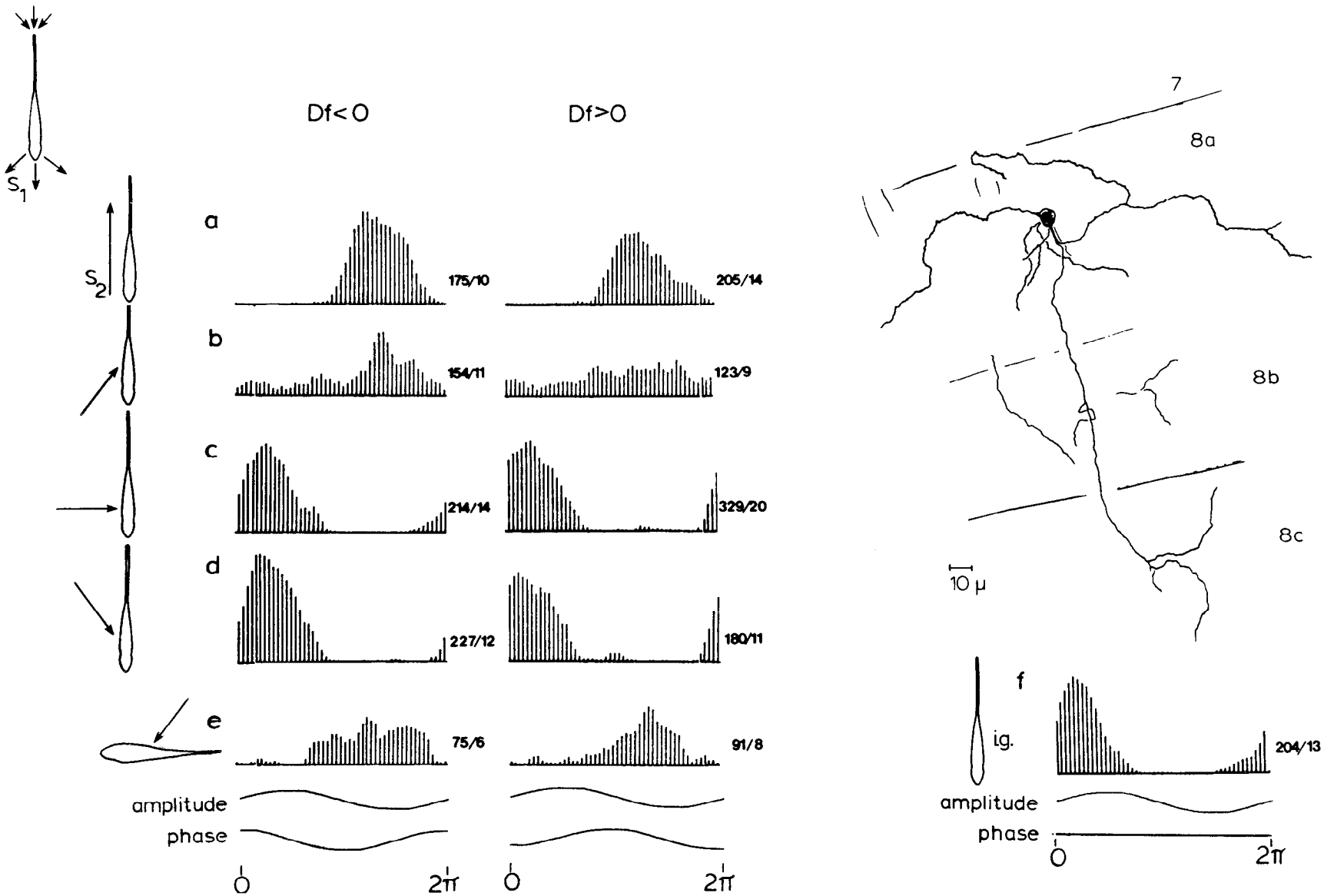

Figure 3. Example of a unit excited only by AMs with the same presentation of data as in Figure 2. Each bin represents 10 msec. This neuron of lamina 8a responded to an increase in stimulus amplitude (*f*, identical geometry (*i.g.*)). The distribution of spikes within the beat cycle was independent of the sign of *Df* regardless of the orientation of the S_2 field (*a* to *e*). Note, however, that the distribution of activity within the beat cycle depends on the orientation of S_2. This is because the angle between the positive current vectors of S_1 (*inset* in *upper left*) and S_2 (*arrows* in *a* to *e*) varies as a function of body location and S_2 orientation. The receptive field of this neuron is presumably located on the left side of the fish's head. In *c* and *d*, the angles between S_1 and S_2 are acute for this region of the body, whereas in *a* and *e* the angles are obtuse. Thus, a maximum in the beat envelope occurs within periods of the beat cycle histogram which are 180° out of phase for these two situations. The amplitude and phase functions below the beat cycle histograms indicate the modulation in amplitude and phase resulting from an electronic addition of S_1 and S_2 and, thus, are representative of the stimulus only at the regions of the body surface where S_1 and S_2 form acute angles. For obtuse angles these functions must be shifted by half a beat cycle. Note, also, that this neuron is driven minimally in *b*, where the direction of the S_2 current is almost parallel to the surface of the left side of the head; with this geometry there is minimal interference between S_1 and S_2 in this region of the body.

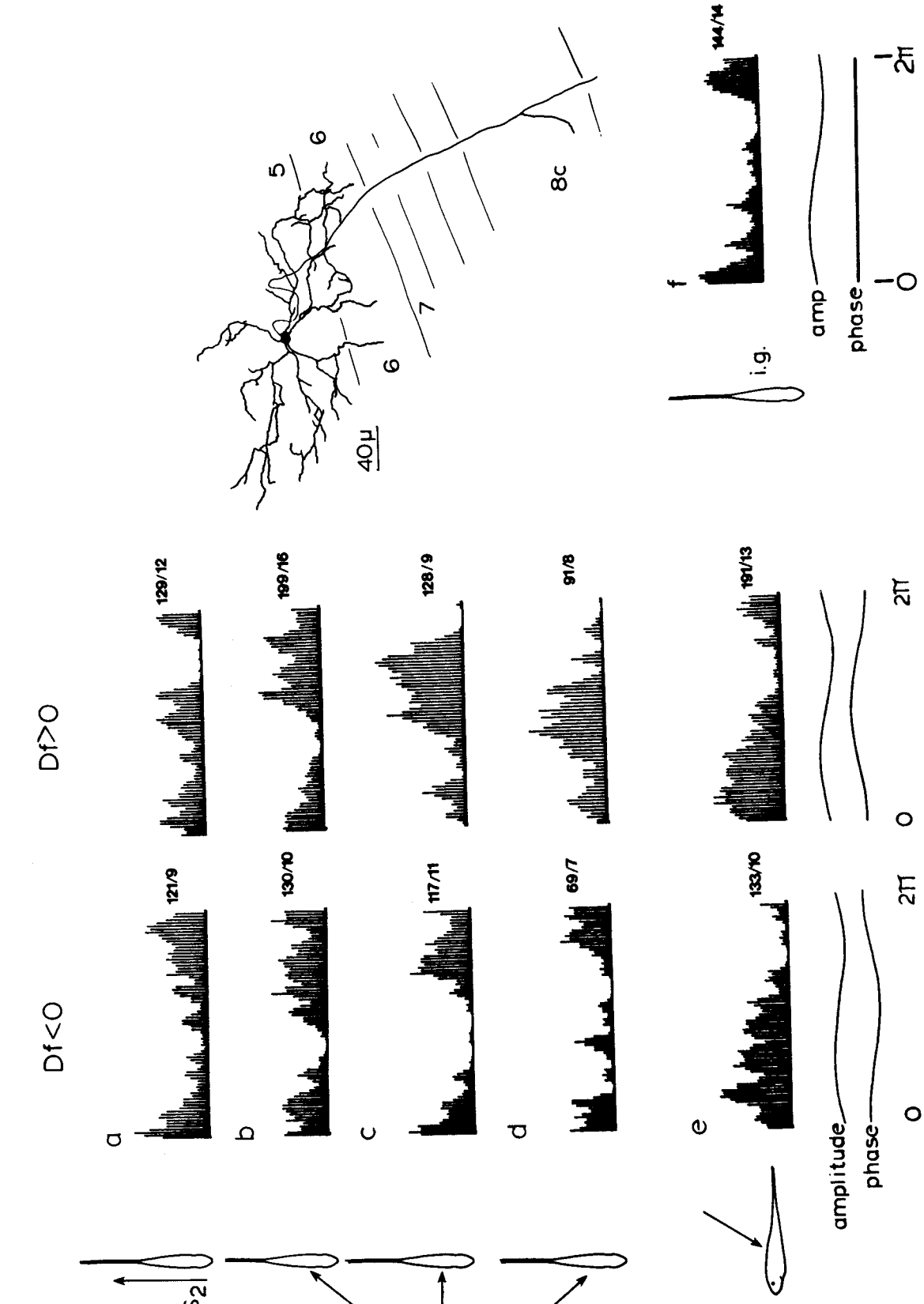

Figure 4. Example of a unit which was excited by increases in stimulus amplitude (*f*) and was also sensitive to differential phase (*c* and *d*). This neuron is a large multipolar cell of lamina 5. The presentation of data is as in Figures 2 and 3. Each bin represents 8 msec.

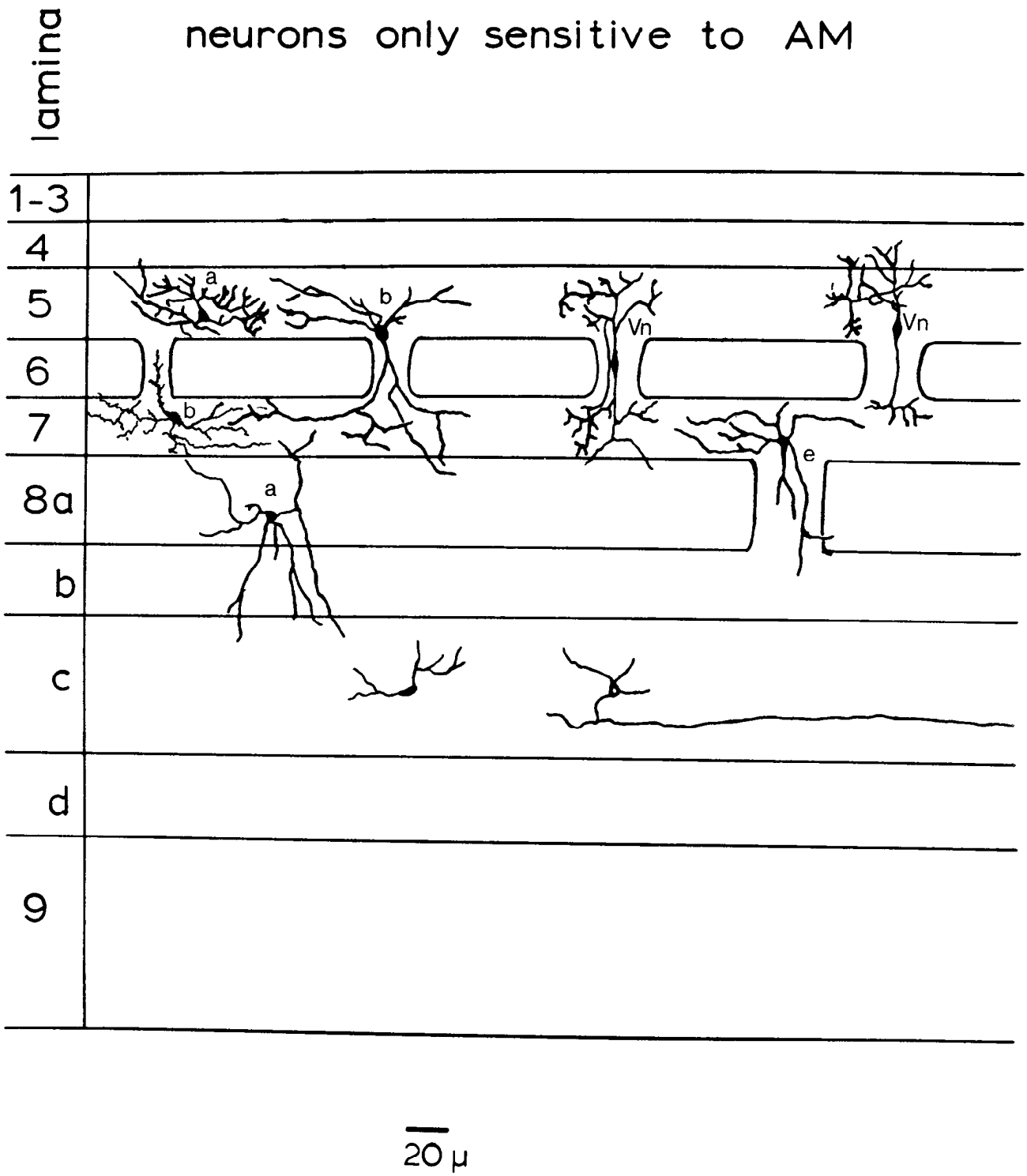

Figure 5. Laminar distribution of cell types which were sensitive to AMs but not to modulations of differential phase. Neuropil regions of lamina 6 (*rectangular areas*) are penetrated by vertical neuropil (*Vn*) bundles which contain afferent and efferent axons as well as a characteristic cell type. *Lowercase lettering* adjacent to each cell refers to a similar cell type identified by Carr and Maler (1985) in Golgi material. Reconstructions were based on camera lucida drawings of cells filled with Lucifer Yellow.

of histograms). For two types, b and c, the following physiological and anatomical profiles were found. Type b is a horizontal cell with a bipolar soma (Fig. 5). This neuron has horizontally distributed, spiny dendrites that are restricted to lamina 7 and, to a lesser extent, to the vertical neuropil zones of lamina 6. The single type-b neuron labeled was excited only by AMs. The type-c neurons are bushy and glioform with a rather small soma (6 to 9 μm; (Fig. 6). Golgi-stained neurons and Lucifer Yellow-filled cells have dendrites which cluster together and project into the neuropil regions of lamina 6. Both Lucifer Yellow-filled cells exhibited sensitivity to differential phase. Complete physiological records have not yet been obtained for the other cell types of lamina 7.

Thus, for neurons which were located in laminae 5 or 7 or in the vertical neuropil regions, sensitivity to modulations of differential phase correlates with the presence of dendritic extensions into lamina 6. For these neurons, AM sensitivity and differential phase sensitivity were quantified according to the Fourier procedures outlined under "Materials and Methods." Larger values reflect greater

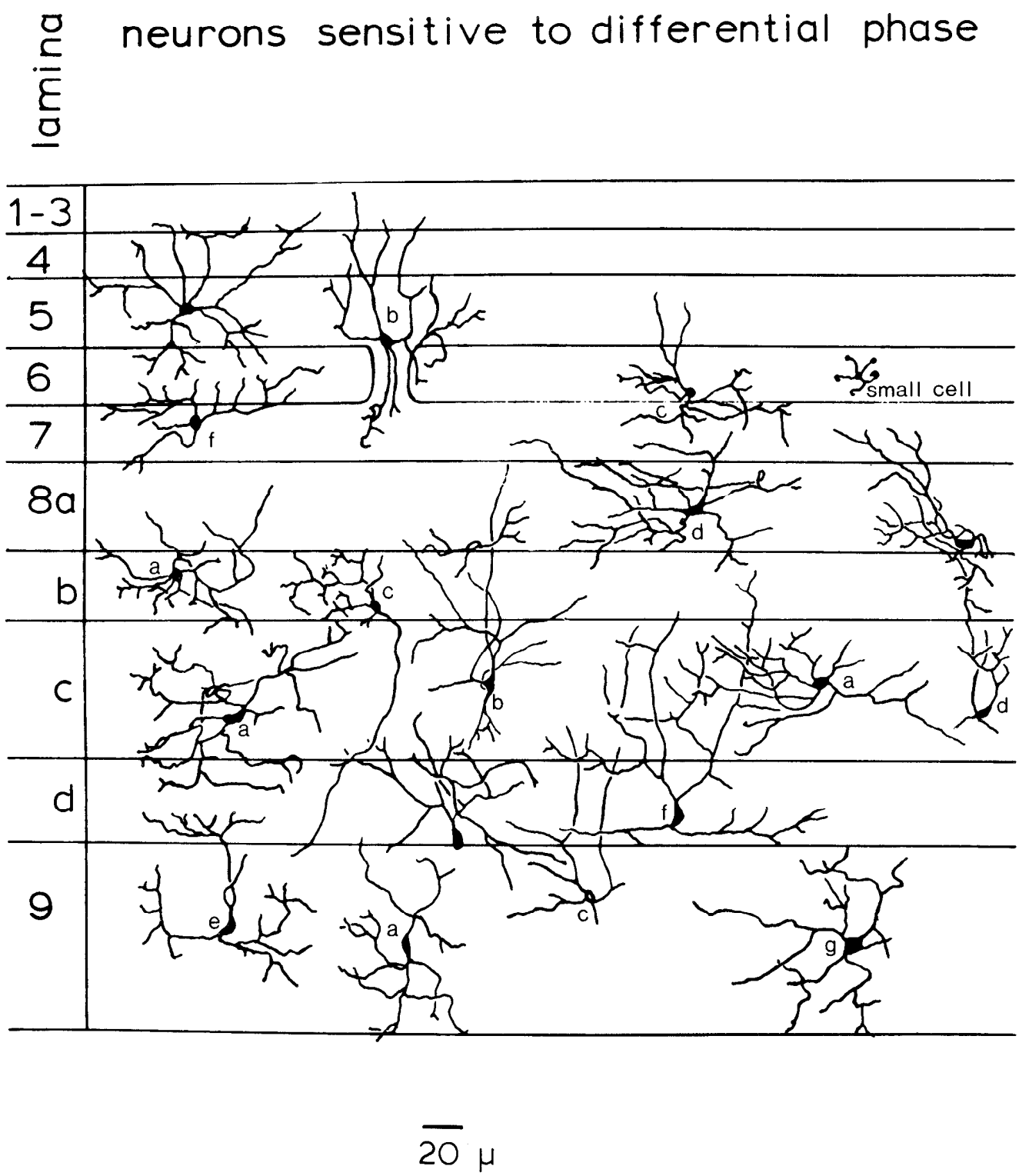

Figure 6. Laminar distribution of neurons which were sensitive to differential phase. As in Figure 5, *lower-case letters* adjacent to cells refer to cell types identified from Golgi material. The lamina 5 cell on the *left* has not yet been identified in Golgi material.

sensitivity. These data are presented in Figure 8. In general, neurons which had dendritic arbors in lamina 6 exhibited strong sensitivity to differential phase, whereas cells which were weakly phase sensitive did not have dendrites in lamina 6.

Interlaminar connections. Carr and Maler's (1985) Golgi analysis revealed 27 cell types in the deeper laminae of the torus (laminae 8a, b, c, d, and 9). Twenty of these cell types have been labeled with Lucifer Yellow. A consistent finding in both types of material is that these neurons do not send dendrites into the neuropil of lamina 6. Since the small cells of lamina 6 appear not to project to other laminae, it is unlikely that the phase sensitivity of neurons in the deeper laminae results from direct input from the small cells of lamina 6. The neurons of lamina 8c could receive differential phase information by way of interlaminar projections from cells of laminae 5 and 7 (Fig. 9). Ten neurons (seven lamina 5 cells and three lamina 7 cells) were labeled sufficiently to reveal an axon in deeper laminae of the torus. Seven of these neurons had axonal collaterals which were confined to lamina 8c. Three of these cells are type-b neurons

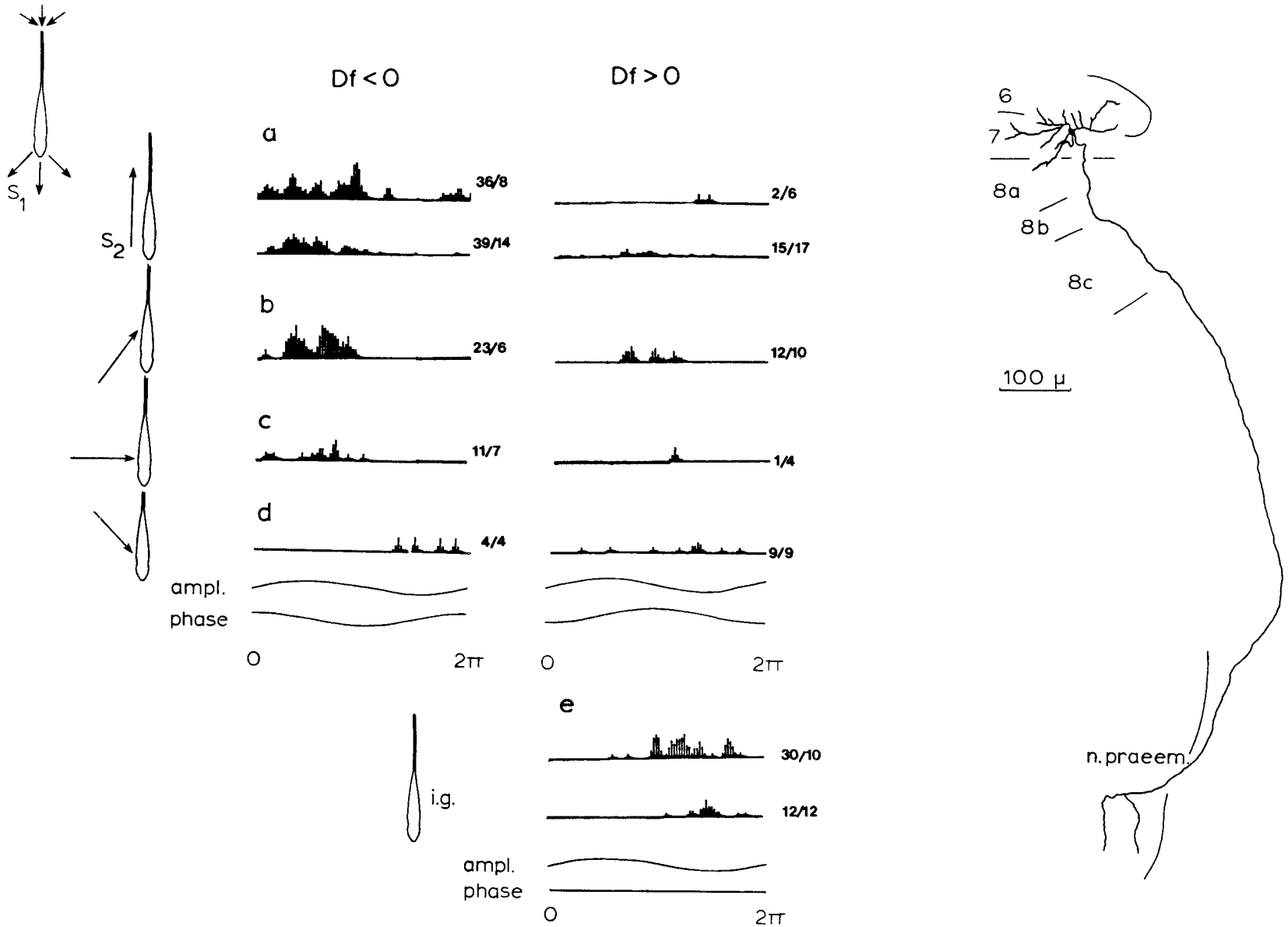

Figure 7. *a*, Horizontal cell (type f) of lamina 7 which exhibited "sign selectivity" (i.e., responded more vigorously for one sign of Df than for the other sign. This unit responded best when the frequency of S_2 was lower than that of S_1 ($Df < 0$). This sign selectivity was most apparent when S_2 was presented longitudinally (*top set* of *histograms*) *b*, The cell responded to a decrease in stimulus amplitude when S_1 and S_2 were presented with identical geometry. Two sets of data are presented in *a* and *e*. Presentation of data is as in Figures 2 to 4; bin width is 6 msec. The camera lucida drawing on the *right* shows that this neuron projects to the nucleus prae-eminentialis.

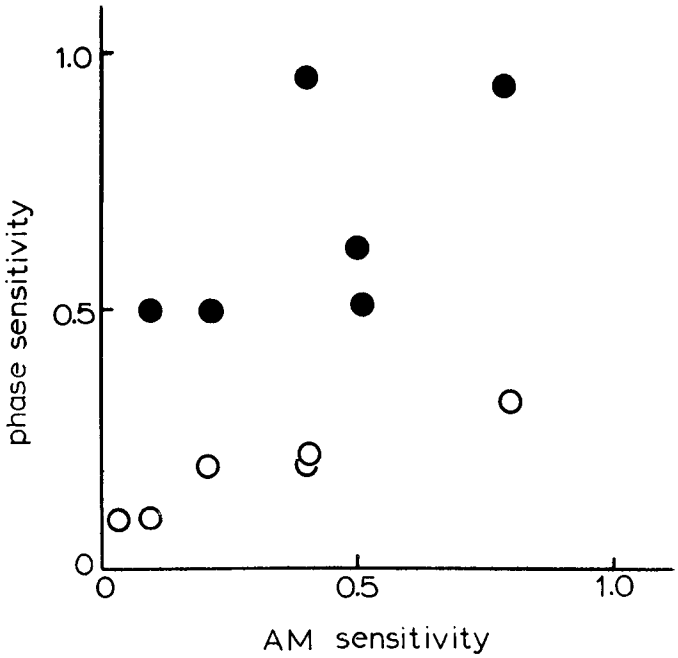

Figure 8. Differential phase sensitivity versus AM sensitivity for toral neurons of laminae 5 and 7 and the vertical neuropil regions connecting these two laminae. Two groups are represented, neurons which had (●) and neurons which did not have (○) dendrites in lamina 6. Larger values represent stronger sensitivity. The measurement of sensitivities is explained under "Materials and Methods." The separation of *solid* and *open circles* along the *ordinate* is significant ($p < 0.001$, Mann-Whitney *U* test).

in lamina 5, one is a large multipolar cell of lamina 5, one is a type-a cell of lamina 5, another is a type-c neuron of lamina 5, and the last is a type-c cell of lamina 7. Only one neuron, a type-a cell of lamina 5, was found to have an arborization in lamina 8a. The other two neurons, a type-f cell of lamina 7 and a type-b cell of lamina 5, exited the torus without forming any obvious collaterals.

Extra-toral projections. Projection neurons (neurons which sent an axon out of the torus) were found in all laminae except 1, 2, and 6. No cells of the first two laminae were labeled. Twenty-five toral neurons were labeled sufficiently strongly to permit identification of their efferent targets. The optic tectum was the most common target of these cells (23 of a total of 25 neurons sent projections there). In contrast, only two neurons were found that projected to the nucleus electrosensorious complex. The importance of this complex for controlling the JAR is indicated by the finding of sign-selective units in this area and by the finding that increases in EOD frequency can be produced by electrical stimulation of the region (Bastian and Yutas, 1984). Projections to the nucleus prae-eminentialis, lateral mesencephalic reticular area, or both, were observed in approximately two-thirds of the cases; seven neurons were found which sent an axon to all three sites. Lamina 8b neurons were unique in that projections to the nucleus prae-eminentialis were never observed (all five cases).

Discussion

The principal findings of this work are as follows. First, the vast majority of the neurons in laminae 8a, b, c, d, and 9 is sensitive to differential phase, whereas purely AM-sensitive cells are often found in laminae 5 and 7 and within the vertical neuropil regions of lamina 6. Second, with regard to the neurons of laminae 5 and 7, differential phase sensitivity is related to the presence of dendritic extensions into lamina 6. Since differential phase is computed by the small cells of lamina 6, this suggests that lamina 5 neurons and lamina 7 neurons that are sensitive to differential phase receive this information via their dendrites in lamina 6. These data parallel earlier work demonstrating a strong relationship between the dendritic structure of pyramidal cells in the ELLL and their function (Saunders and Bastian, 1984). Similarly, functional differentiation can be related to morphological distinctions in the case of X-, Y-, and W-cells of the A and C laminae of the cat lateral geniculate nucleus (Friedlander et al., 1979, 1981; Stanford et al., 1981). The dendritic arbor of X-cells is confined to lamina A, whereas the dendrites of Y-cells always cross the laminar borders.

These findings support the hypothesis that, in laminated structures, the functional differentiation of a neuron may, in part, be a consequence of the laminar location of its dendrites; each neuron is presumed to receive synaptic input from all afferents that have terminations in its dendritic zone (Maler, 1976). Since laminated structures are characterized by a spatially discrete pattern of afferent innervation, specific functional cell types could result from differential sampling of these inputs. The dendritic structure of a neuron, therefore, could determine its functional integration.

Physiological and anatomical studies of the visual cortex have provided further evidence in support of this "laminar specificity of function" hypothesis. In the striate cortex of the cat, for example, only those cells receiving direct input from X- and Y-afferents of the lateral geniculate nucleus have simple receptive field properties (Gilbert, 1977; Gilbert and Wiesel, 1979). Furthermore, the specific functional properties of these simple cells are related to the afferent zone in which they are found: X-afferents terminate in lamina 4c, whereas Y-afferents are found in lamina 4ab.

These results are strikingly similar to those obtained from studies of the torus semicircularis of *Eigenmannia*. In this structure, phase-coding afferents terminate exclusively in lamina 6 (Carr et al., 1981). Correspondingly, the giant neurons, which are restricted to this lamina, are the only phase-coding cells in the torus. The small cells, which also are restricted to this zone, are the sole recipients of input from the giant neurons and uniquely compute differential phase information (Heiligenberg and Rose, 1985).

In addition to their termination in the simple cell zones, X- and Y-afferents to the cat striate cortex also appear to project to lamina 5 (see review by Gilbert, 1983). Similarly, pyramidal cell afferents from the ELLL have terminations in laminae 8c and 8d of the torus, in addition to the more substantial input to laminae 5 and 7 (Carr et al., 1981; Heiligenberg and Rose, 1985).

To understand the processing of sensory information in laminated structures, it is important to determine the relative functional consequences of the laminar segregation of inputs to the various dendritic fields of a particular neuron. As in cortical structures of mammals, many neurons of the torus of *Eigenmannia* distribute their dendrites among several laminae. The present work demonstrates, for the neurons of laminae 5 and 7, a functional relationship between the laminar organization of a neuron's dendrites and its function.

Two lines of evidence suggest that the small cells of lamina 6 do not project outside this lamina. First, injections of horseradish peroxidase (HRP) into laminae outside of lamina 6 have failed to back-label these small cells. Furthermore, small cells which were labeled by intracellular iontophoresis of Lucifer Yellow did not reveal an axon leaving lamina 6. Nevertheless, it is conceivable that our techniques have been unable to reveal interlaminar projections of these small cells. The neurons of laminae 5 and 7 have axonal collaterals in lamina 8c, thus providing one mechanism by which differential phase information can reach the deeper laminae of the torus.

The finding that lamina 8b neurons do not send a projection to the nucleus prae-eminentialis is consistent with earlier results based on retrograde transport of HRP (Carr et al., 1981). The eurydendroid cells of the lobus caudalis (which project to lamina 8b) receive indirect input from the torus through afferents from the nucleus prae-eminentialis. It appears, therefore, that the neurons of lamina 8b do not send information back to the cerebellum.

The most common target of the toral efferents is the optic tectum. In all cases these projections terminate in the deep layers of the tectum, most commonly the stratum album centrale, but sometimes extending into the stratum griseum and fibrosum centrale. This

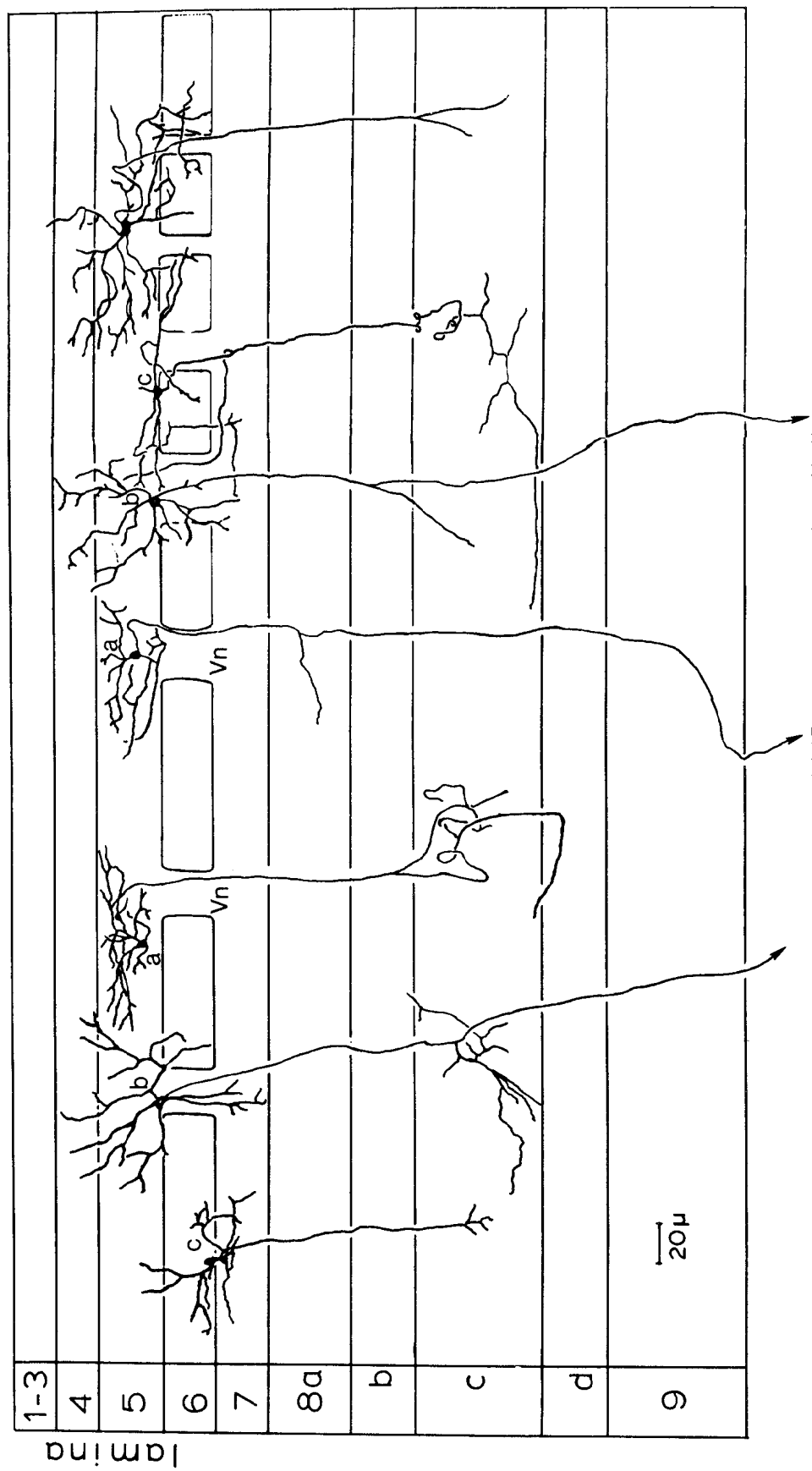

Figure 9. Camera lucida drawings of neurons of laminae 5 and 7 which had axonal collaterals in the deep laminae of the torus. Three type-b cells of lamina 5 had collaterals in lamina 8c; two of these are shown. The axons of lamina 5 neurons travel through the columns of vertical neuropil (*Vn*). The neuron type on the *far right* has not yet been observed in Golgi studies. The Golgi type of all other cells is indicated by the *lower-case letter* adjacent to the cell.

electrosensory input is consistent with the multimodal nature of the tectum observed in many species of animals (Gordon, 1973; Dräger and Hubel, 1976; Newman and Hartline, 1981). Whereas visual input terminates in the superficial layers, other sensory modalities are restricted to the deeper layers (Casagrande et al., 1972). Bastian (1982) has demonstrated that tectal neurons respond to electric images produced by moving objects, many being bimodal and having visual and electrical receptive fields in spatial register.

The two types of pyramidal cells found in the ELLL are functionally distinct. Basilar pyramidal neurons are excited by increases in stimulus amplitude, whereas nonbasilar pyramidal cells respond to decreases in stimulus amplitude (Saunders and Bastian, 1984). It was of interest, therefore, to determine whether these two AM-processing "channels" remain discrete at the level of the torus. Whereas most AM-sensitive neurons were either E type (excited by increases in stimulus amplitude) or I type (driven by decreases in stimulus amplitude), these functional classes did not correspond to morphologically distinct cell types. This conclusion is not entirely surprising, given that the projections from the basilar and nonbasilar pyramidal neurons of the ELLL terminate in the same regions of the torus (Heiligenberg and Rose, 1985). Basilar pyramidal cells (E-units) sample primary afferent information directly via their basilar dendrite. Nonbasilar pyramidal neurons (I-units) receive this information through inhibitory interneurons, being inhibited, therefore, by an increase in stimulus amplitude (Maler et al., 1981). The morphological distinction between these two types of pyramidal cells thus appears to be causally related to their physiological differences. Since E and I functions are not generated at the torus and are only relayed on to other targets, there is no reason to expect that E-cells and I-cells of the torus should be morphologically distinct. These observations suggest the following hypothesis. Functional differentiation of neurons need not be associated with morphological differentiation if their response selectivity already exists in the neurons presynaptic to them. Morphologically different neurons, however, should be functionally different.

The finding that many morphological classes of toral neurons can be classified as either E type or I type suggests that there is little connectional specificity between pyramidal afferents of the ELLL and toral cell types. Since few E-I units are found, one may speculate that, during development, a toral neuron receives synaptic input from a pyramidal cell of the ELLL and then becomes refractory to additional innervation from this structure. This process may be similar to afferent-selecting mechanisms which occur in the visual system (see review by Sherman and Spear, 1982).

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