MICROCIRCUITRY OF BipOLAR CELLS IN CAT Retina1
BARBARA A. McGUIRE,2 JOHN K. STEVENS,3 AND PETER STERLING

Department of Anatomy, School of Medicine, University of Pennsylvania, Philadelphia, Pennsylvania 19104

Received October 31, 1983; Revised June 4, 1984; Accepted June 8, 1984

Abstract
We have studied 15 bipolar neurons from a small patch (14 × 120 μm) of adult cat retina located within the area centralis. From electron micrographs of 100 serial ultrathin sections, the axon of each bipolar cell was substantially reconstructed with its synaptic inputs and outputs by means of a computer-controlled reconstruction system. Based on differences in stratification, cytoarchitecture, and synaptic connections, we identified eight different cell types among the group of 16 neurons: one type of rod bipolar and seven types of cone bipolar neurons. These types correspond to those identified by the Golgi method and by intracellular recording. Those bipolar cell types for which we reconstructed three or four examples were extremely rare in fovea, area centralis, and cytoarchitecture, and also in the quantitative details of their synaptic connections. They appeared quite as specific in these respects as invertebrate "identified" neurons.

The synaptic patterns observed for each type of bipolar neuron were complex but may be summarized as follows: the rod bipolar axon ended in sublamina b of the inner plexiform layer and provided major input to the AII amacrine cell. The axons of three types of cone bipolar cells also terminated in sublamina b and provided contacts to dendrites of on-β and other ganglion cells. All three types, but especially the CbA and CbB, received gap junction contacts from the AII amacrine cell. Axons of four types of cone bipolar cells terminated in sublamina b of the inner plexiform layer and contacted dendrites of off-β and other ganglion cells. One of these cone bipolar cell types, CbA, made reciprocal chemical contacts with the bipolar appendages of the AII amacrine cell.

These results show that the pattern of cone bipolar cell input to β (X) and probably α (Y) ganglion cells is substantially more complex than had been suspected. At least two types of cone bipolar could be each type of ganglion cell where only a single type had been anticipated. In addition, many of the cone bipolar cell pathways in the inner plexiform layer are available to the rod systems, since at least four types of cone bipolar receive electrical or chemical inputs from the AII amacrine cell. This may help to explain why, in a retina where rods far outnumber the cones, there should be so many types of cone bipolar cells.

Bipolar cells are the only retinal neurons directly interposed between photoreceptors and ganglion cells. Therefore, knowledge of their connections is likely to be one key to understanding the basis for the receptive field properties of retinal ganglion cells. Until recently, it was thought that bipolar cell anatomy and physiology in the cat retina were relatively simple, because only three kinds of cone bipolar cells were known. Boycott and Kolb (1973) had observed one morphological variety of cone bipolar with dendrites invaginating the cone pedicle and axon branching in sublamina b of the inner plexiform layer. They observed two other morphological forms of cone bipolar with dendrites in "flat" contact with the basal surface of the cone pedicle and an axon, either "smooth" or "vesicular," branching in sublamina a. The physiological interpretations at the time were correspondingly simple: the imaging cone bipolar was thought to depolarize at light on and excite the on-center ganglion cells, and the flat cone bipolar was thought to hyperpolarize at light on and to excite (at light off) the off-center ganglion cells (Famiglietti and Kolb, 1979; Nelson et al., 1978).

Several lines of evidence now suggest that the situation is more complicated. In most species, there are many different kinds of bipolar cell (e.g., Kulwin, 1979; Rome, 1977, 1979; Wilkoch and Stell, 1979, West, 1978). In the cat, in particular, there seem to be at least eight morphological forms of cone bipolar neurites based on studies of Golgi-stained retinas (Famiglietti, 1981; Kolb et al., 1981). This diversity suggests that the circuits involving bipolar cells may be more complex. Some bipolar cells have been shown to accumulate glycine (Nakamura et al., 1978; Pourcho, 1980), which is thought to act as an inhibitory transmitter in the retina (Wyatt and Daw, 1976; Miller et al., 1977). In addition, both depolarizing and hyperpolarizing types of cone bipolar cells seem to send axons to a single sublamina of the inner plexiform layer where they

---

1 This work was supported by National Institutes of Health Grants EY03382, EY04903, EY01585, and Research Career Development Award EY00486 (to P. S.). During part of this work, J. R. S. was supported by a Canadian Medical Research Council grant. We thank Neil Fadda and Robert G. Smith for their constant assistance, Fred Lettieri, Ed Shain, and Alfred M. Heymann for technical help, Donna Henney, John Magli, and Dalby Natale for technical help, and Susan Heemiller for help with the illustrations. Thanks also to Michael Freed and Dr. James Hinnis for useful discussions.

2 To whom correspondence should be sent, at her present address: The Rockefeller University, 1220 York Avenue, New York, NY 10021.

3 Present address: Department of Physiology, Presbyterian Memorial Neurosciences Unit, Toronto Western Hospital, 399 Bathurst Street, Toronto, Ontario M5T 2C8 Canada.
Figure 1. Low magnification, electron micrograph of sections 42 to 46. The rectangle in the second section indicates the six regions photographed per section. Ganglion cells turned a solid red in this region.

Figure 2. Low magnification electron micrograph of section 59. Numbers mark the assigned seven reconstructed bipolar cells: 1, on-3 cell; BV, blood vessel; GCL, ganglion cell layer; INL, inner nuclear layer; IPL, inner plexiform layer; OPL, outer plexiform layer.

Figure 3. Schematic diagram of right eye apex temporal area. Tissues for the sets were located ED about 2° (0.4 to 0.5 mm) from the center of the area centralis. O.D., optic disk.

can contact ganglion cells of rod type (Kolb and Nelson, 1983; Nelson and Kolb, 1983). In view of these growing complexities, it seemed critical to develop a comprehensive classification of bipolar neurons in which each of the diverse morphological categories could be associated with a distinctive cytoarchitecture, synapses, and chemistry. If such a classification could be developed, it would then be possible to determine the patterns of contact made by each bipolar cell type with specific types of ganglion cells.

Nelson and Kolb (1983) have approached this problem by recording the responses of bipolar neurons, injecting horseradish peroxidase, and observing the filled cells in the light and electron microscopes. They have established that certain cone bipolar cells have characteristic physiological responses and characteristic synaptic connections. The symmetric patterns were not determined quantitatively, though, for any type. Also, since the method requires accumulating cells at different eccentricities and from different retinas, it is unknown how regular the features of a particular type might be and whether all types are present at every point in the retina.

We have taken a complementary approach, that of reconstructing a group of 15 bipolar neurons from serial electron micrographs taken within a small region of a single retina. The patterns of synaptic contact formed by bipolar neurons of specific morphology and cytoarchitecture were studied qualitatively. The associations we observed among morphology, cytoarchitecture, and synaptic patterns permitted us to see the group of 15 neurons in eight types. In the absence of variation due to differences in eccentricity and differences between animals, the individual members of certain types were remarkably similar in morphological and synaptic cytoarchitecture. The bipolar cell types described here match rather well the categories described by Guldberg (1980; Kolb et al., 1981) and by intracellular recording followed by injection of horseradish peroxidase (Kolb and Nelson, 1983; Nelson and Kolb, 1983). The resulting classification scheme for bipolar cell types has formed the basis for subsequent study of the connections of specific types of bipolar neurons with other neuronal elements (J. A. McGuire, J. K. Stevens, and P. Sterling, manuscript in preparation).

Materials and Methods
Tissue preparation and photography. In a normal adult rat anaesthetized with Nembutal, the area of the eye was slit and the animal was perfused with a mixture of 15% glutaraldehyde/15% paraformaldehyde.
Figure 4. Computer reconstructions of the 15 bipolar cells placed as they appeared in the series. Shaded cells have darker cytoplasm. The numbers represent RB cells 1, 8, 9; CB₁ cells 2, 6, 10, and 11; CB₂, cell 4; R₈, cell 3; R₁₂, cells 1 and 14; CB₁₂a, cells 5 and 15; CB₁₂b, cell 12; and CB₁₃, cell 7. DNL, distal cell layer; INL, inner nuclear layer; IPL₁, inner plexiform layer; GFL, outer plexiform layer.

Table 1

Morphological characteristics of subtypes of bipolar cells

<table>
<thead>
<tr>
<th></th>
<th>RB₁</th>
<th>RB₂</th>
<th>CB₁</th>
<th>CB₁₂a</th>
<th>CB₁₂b</th>
</tr>
</thead>
<tbody>
<tr>
<td>Dendritic diameter (μm)</td>
<td>6.2</td>
<td>6.5</td>
<td>7.6</td>
<td>8.7</td>
<td>7.9</td>
</tr>
<tr>
<td>Soma position (μm from IPL₁)</td>
<td>11.5</td>
<td>12.1</td>
<td>12.2</td>
<td>12.2</td>
<td>12.2</td>
</tr>
<tr>
<td>Primary dendritic diameter (μm)</td>
<td>1.3</td>
<td>2.0</td>
<td>1.6</td>
<td>1.6</td>
<td>2.6</td>
</tr>
<tr>
<td>Primary axon diameter (μm)</td>
<td>1.1</td>
<td>1.2</td>
<td>1.2</td>
<td>1.6</td>
<td>1.4</td>
</tr>
<tr>
<td>Axon contour</td>
<td>Vertical, scalloped</td>
<td>Widens, SS-4</td>
<td>Widens, SS-5, exp. 4.5</td>
<td>No widening</td>
<td>No widening</td>
</tr>
<tr>
<td>Axon stratification</td>
<td>SS-5</td>
<td>SS-5, exp. 4.5</td>
<td>SS-5</td>
<td>S₁</td>
<td>S₁, S₄</td>
</tr>
<tr>
<td>Conglomeration density</td>
<td>Very dark</td>
<td>Pale</td>
<td>Medium</td>
<td>Medium</td>
<td></td>
</tr>
</tbody>
</table>

* Cell type.
* Cell number.
* Measurements unavailable due to incomplete reconstruction.

(200 cell) followed by 2% glutaraldehyde/2% paraformaldehyde (800 ml). The aldehyde solutions were dissolved in 0.12 M phosphate buffer at a pH of 7.4. The next day the tissue was osmicated, stained in bloc with uranyl acetate (4% in sodium acetate buffer), dehydrated through a series of methanols, and embedded in Epon. We cut a series of 100 μm thick sections (estimated 0.075 μm thick) on a Leica MT 250 microtome, mounting the sections on 0.25 μm slides and staining further with uranyl acetate and lead citrate. Each section was photographed on an EOCLE 1000 microscope at ×200 kV. No overlapping micrographs were taken of each section (Fig. 1a) at about 1000 to cover an area of about 100 μm × 100 μm (Fig. 1b). Reconstruction. Bipolar cells were reconstructed using a computer-assisted video system designed for rapid viewing of corresponding areas in adjacent sections and for easy digitization of cell profiles. This system is described in detail by Steves et al. (1986). Briefly, the EM images of a particular area were roughly aligned and copied onto a thin film strip which was viewed through a microscope and displayed on a microprocessor controlled, high resolution video monitor. The profiles of a bipolar cell were followed within a section by moving the filmstrip laterally (X, Y) by means of stepping motors and between sections by advancing the filmstrip. The X,Y coordinates within a section and section number for each profile were recorded on the computer. We aligned the corresponding profiles in adjacent sections using small structures such as microvilli as fiducials, and then digitized all of the profiles. Also recorded were the positions and identities of all presynaptic and postsynaptic profiles. The data were transferred to a PDP 11/34 computer which "stacked" the profiles in register and removed hidden lines to provide an image of the whole cell. The illustrations display every synaptic contact, rather than "leaving" the ones which are on the rear surface of the cells. The reconstruction procedure appears to represent quite faithfully the actual shape of these neurons because they were striking similarities between our reconstructions and the morphology of particular bipolar cells described by Görgi innervation.
Bipolar Cell Microcircuity

Fifteen bipolar cell axons were partially reconstructed from a strip of retina 14 × 120 μm. In this region, the cells in the ganglion cell layer formed a solid tile (from one to two cells thick; see Fig. 1). There were 10,000 cells/mm² which placed this patch of retina within the area centralis, about 2′ from its center (unpublished results; see Fig. 3). Although we focused on characterizing the axonal arbors, for 12 cells the soma was also reconstructed; and for two dendrites, reconstruc-
ted to their tips (Fig. 4). Particular attention was devoted to identifying all of the synaptic contacts made by each bipolar cell axon and to determining the strata of the inner plexiform layer (SI to S5) in which each arborized. Because we recon-
structed a small portion of the bipolar cells in this strip of retina (perhaps 5% to 20% of the total), it was not possible to determine the relative abundance of any one type.

Profiles in the inner plexiform layer were identified according to previously published criteria (see, for example, Dowling and Boycott, 1986; Kolb, 1979; Stevens et al., 1980). Thus, premyn-
gap profiles with synaptic vesicles and synaptic ribbons were called "bipolar," those with vesicles and making conventional contacts were called "amacrines." Amacrine cells postsynaptic and postsynaptic to the same bipolar cell (even if not in the same section) were called "reciprocal." Postsynaptic profiles lacking vesicles but containing microtubules and clusters of ribonucleosides were called "ganglion cell dendrites." Ganglion cell dendrites with dark cytoplasm always belonged, when traced to a soma, to a β cell. The largest dendrites with pale cytoplasm were probably from α cells (Stevens et al., 1980). In the outer plexiform layer, pale profiles with synaptic vesicles and con-
ventional contacts onto bipolar cell processes are believed to be largely from interplexiform cells (Kolb and West, 1977; Nakakura et al., 1980), and we have referred to them that way.

Rod bipolar cells

The soma of these rod bipolar cells (RBs) were reconstructed in their entirety. These were not traced to their connections with the rod spherules but were identified by comparison to previous descriptions (Kolb and Pasquetti, 1974; Kolb, 1979). The rod bipolar cell somata were 6.3 to 7.6 μm in diameter (Table 1), smaller than those of cone bipolar neurons, and profuse with a single gradually tapering dendrite ascending
toward the outer plexiform layer. Their cytoplasm was distinc-
tively dark (as in Fig. 5), but not so dark as the cytoplasm of Müller cells. The axons, curving slightly, descended through sublaminas a without branching. Upon reaching sublaminas b (straturn 3), each axon thinned somewhat, assumed a scal-
lipped contour, and began to make many synaptic contacts (Figs. 6 and 7). The frequency of these contacts increased as the rod bipolar cell axons passed through strata 4 and 5 where, at the edge of the ganglion cell layer, the axons terminated (Fig. 6).

The three rod bipolar cell axons received, respectively, 35, 39, and 42 inputs from amacrine cells (Table II). In each case almost exactly 70% (69%, 69%, 71%) of the contacts were from reciprocal amacrine cell varicosities, the remainder being from nonreciprocal profiles. Although all of these postsynaptic varicose-
ous contained one another, it is known that the reciprocal contacts derive at least from one specific type of amacrine (Kolb and Nelson, 1983), all of which accumulate GABA (Fréé and Sterling, 1980; Pochro and Geisel, 1983) and possibly indoi-
leamine (Holmgren-Taylor, 1982).

The rod bipolar neurons provided outputs to, respectively, 27, 28, and 52 synaptic ribbons. Almost invariably a ribbon was presynaptic to two processes (Table III) and thus formed the classic "dyad" arrangement described by Kjaeld (1960), Dowling and Boycott (1966), and Kolb (1979). In virtually every case one member of the dyad belonged to a reciprocal amacrine cell process. The other member, when it could be identified, always belonged to an All amacrine cell. That of the 65 ribbons contacts where both postsynaptic profiles were identified, 64 were onto a pair of amacrine cell profiles. At 23 of the 87 dyads examined, the nonreciprocal postsynaptic process occurred in the slender portion of All cell processes but was lost before the identification was secure. In the best case, however, where 94% of the postsynaptic profiles were identified, every postsynaptic process belonged to an amacrine cell, 46% being reciprocal and 48% being All. As a rule, therefore, each rod bipolar cell ribbon synapse was made with a pair of amacrine cell processes: the All process, and a varicosity which in every case returned a reciprocal synapse to a rod bipolar cell occasionally contacted ganglion cell dendrites. Cell 7, for example, directed one ribbon contact to a pale ganglion cell dendrite (Fig. 7A and 7B). Two other rod bipolar (not reconstructed) each provided one contact to another pale ganglion cell dendrite (Fig. 7A and 7B). Both rod bipolar cells were All. However, direct rod bipolar to-ganglion cell contacts were known to exist in rabbit retina (Raviss and Ravish, 1967), this is the first report of such synapses in the cat.

The three reconstructed rod bipolar cells differed somewhat in the proportion of contacts in each of the three layers and in the form of the soma and proximal dendrite, but for other features they exhibited remarkable constancy. Their cy-
tology and axon morphology were indistinguishable. Thus, the depth in the inner plexiform layer at which the axons began to make synaptic connections to within 25 μm and the synaptic patterns of the three axons were virtually identical.

Conc bipolar cells arborizing in sublaminas a

Six cone bipolar cells with axons arborizing in sublaminas a were partially reconstructed. Three proved to be of three types, which we have called C1a, C1b, and C2b.

C1a. There were four examples of this type, in which cell 11 was most complete (Fig. 8). The soma of cell 11, located high in the inner nuclear layer, was oval (8.5 × 11.2 μm) with pale cytoplasm. Fine processes extended from the upper pole of the soma to ramify among the cone pedicles, and the frequency of the contacts with cones could not be determined. There were two synaptic contacts on these dendrites from the interplexiform cell axon. The axon descended as a single, pale process (about 1.4 μm
Figure 6. Rod bipolar (RB) reconstructions. At the bottom, axons rotated 90° about the X axis with the first and last section numbers indicated. The sublamina vb border was marked in sections by the presence of rod ribbons. Apparatus above the border (top one-third), and rod bipolar ribbon synapses below (bottom two-thirds). Strata 1 and 2 were each half of a, and strata 3 to 5 were each one-third of a. The slender axons pass through sublamina a and then widen and become whithelined upon entering sublamina b where they receive inputs (A) and make outputs (E). INL, inner nuclear layer; IPL, inner plexiform layer; OPL, outer plexiform layer.
In diameter through sublamina a without making any synaptic contacts. Upon entering sublamina b, the axon swelled to 3.5 μm and began to make synaptic contacts within the neuropil (Fig. 3A). In stratum 4 it emitted several thin processes that descended into stratum 5. The other three CBs’ axons were also pale and vertically oriented through strata 4 to 6. Ten percent of the contacts of these bipolar cells were made in stratum 3, 60% in stratum 4, and 30% in stratum 6. The endings of these CBs’ axons and also some of the other types of bipolar cells in sublamina b were invaginated by fine tangential cell and amacr ine cell processes (Fig. 3A and B); none of the bipolar cell axons in sublamina a bore such invaginations.

The CBs, axon terminals received many chemical synaptic contacts from amacrine cells. The most extensively reconstructed axon, that of cell D, received 61 such contacts. Three-fourths of these were from nonreciprocal amacrine cells, i.e., those that provide input to the bipolar cell but do not in turn receive it. The rest of the contacts, roughly one-fourth, were from reciprocal amacrine cell terminals, which, as dyad members, received ribbon-related contacts from the bipolar cell. CBs, cone bipolar cell axons received numerous extensive gap junction contact, mainly within strata 4 and 5, from the processes of all amacrine cells. Cells 6 and 11 each received six such contacts, whereas cells 3 and 10, which were less extensively reconstructed, received three apices. Since most, and perhaps all, of the gap junctions on cell 11 were with the branches of a single A1 cell, there was evidence for very little divergence in the connectivity between A1 and CBs’ cells.

The CBs’ axonal terminals provided a number of synaptic ribbon profiles to numerous post synaptic profiles. There were 88 ribbon contacts from the most extensively reconstructed axon (cell 6) and no fewer than 75 ribbon contacts for the others. Of the profiles that could be identified as being post synaptic to CBs, neurons 60% belonged to amacrine cells and 60% belonged to ganglion cells. Of the 53 ribbon synapses from cell 6 in which both members of the post synaptic dyad could be identified, 41 were onto a ganglion/amacrine cell pair, 11 were onto a ganglion/ganglion cell pair, and 1 was onto a amacrine/amacrine cell pair. Combining the data from all four CBs, axon, 128 dyads were ganglion/amacrine; 42 were ganglion/ganglion; and 1 was amacrine/amacrine. About one-third of the amacrine cell profiles were reciprocal to the CBs’ axon, but the remainder received input without reciprocating. CBs’ cells contacted at least three morphologically different amacrine cell profiles. One kind was a 1- to 2-μm-wide varicosity which consisted of very thin necks at each end. A second was a plexus, oriented radially in sublamina b, and less varicose, with swellings 2 μm at most in diameter, the connections between these varicosities were large and thus easier to follow than the fast type, allowing us in one case to follow about half a dozen interconnected varicosities which received 9 ribbon contacts from a single bipolar cell. The third amacrine cell variety, which received 6 contacts from one CBs’ cell, was a straight, thin (0.5 to 1.0 μm in diameter) process traveling tangentially within strata 3, making occasional punctate synapses onto very fine post synaptic profiles 0.5 to 1.5 μm in diameter. The ganglion cell dendrite receiving CBs’ input were divided about evenly between dark and pale varicosities.

The axons of CBs showed the same regularities of features as those of the rod bipolar cells. All four CBs’ axons swelled and commenced to make synapses at the same depth in stratum 3. Their cytoplasm was pale, and the form of their arborisations in strata 4 and 5 was similar, as were, in quantitative detail, their patterns of synaptic connection. Such evidence of regularity within two types suggested that if any two bipolar cells differ appreciably, they are likely to represent different types. This reasoning led us to distinguish two additional types of cone bipolar cell axons in sublamina b, even though only a single example of each was reconstructed. CBs. The soma of cell 4 was smaller in diameter (7.5 μm) than that of any of the other cone bipolar cells and was located in the middle of the inner nuclear layer (Fig. 10). It was parynomorphic with a single densely packed (5 μm in diameter) tapering gradually toward the outer plexiform layer (Fig. 10). The cytoplasm was medium-dense (Fig. 9). The axon was darker than those of adjacent CBs’ cells but paler than those of neighboring rod bipolars. From the middle of stratum 2 to the middle of stratum 3, the axon gradually thickened to about 2.2 μm in diameter. In the middle of stratum 3, it divided into three branches that arborized in lower stratum 3 and upper stratum 4 where most of the synaptic contacts were located.
TABLE II

Inputs to bipolar cells of sublamina b

<table>
<thead>
<tr>
<th>Source of inputs</th>
<th>RBb</th>
<th>CBBb</th>
<th>CBBc</th>
<th>CBBd</th>
</tr>
</thead>
<tbody>
<tr>
<td>All amacrine</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Reciprocal amacrine</td>
<td>69%</td>
<td>71%</td>
<td>60%</td>
<td>54%</td>
</tr>
<tr>
<td>Neuroepithelial area</td>
<td>31%</td>
<td>29%</td>
<td>31%</td>
<td>76%</td>
</tr>
<tr>
<td>retinal</td>
<td>100</td>
<td>100</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Total no. of inputs</td>
<td>39</td>
<td>53</td>
<td>62</td>
<td>54</td>
</tr>
<tr>
<td>All gap junctions</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

*Cell type.
*Cell number.

TABLE III

Outputs from bipolar cells of sublamina b

<table>
<thead>
<tr>
<th>Type of bipolar cell</th>
<th>CBBb</th>
<th>CBBc</th>
<th>CBBd</th>
</tr>
</thead>
<tbody>
<tr>
<td>All amacrine</td>
<td>25%</td>
<td>25%</td>
<td>25%</td>
</tr>
<tr>
<td>Reciprocal amacrine</td>
<td>50%</td>
<td>50%</td>
<td>50%</td>
</tr>
<tr>
<td>Neuroepithelial area</td>
<td>0%</td>
<td>0%</td>
<td>0%</td>
</tr>
<tr>
<td>Total</td>
<td>100%</td>
<td>100%</td>
<td>100%</td>
</tr>
<tr>
<td>Total no. of axons</td>
<td>27</td>
<td>28</td>
<td>32</td>
</tr>
</tbody>
</table>

*Cell type.
*Cell number.

Cell 4 received 54 chemical contacts from amacrine cell profiles, which were almost equally divided between reciprocal ones which also received ribbon inputs from cell 4 and nonreciprocal ones which did not. This cell bipolar cell also received a single, small gap junction from an All amacrine cell. Cell 4 provided 57 ribbon contacts to postsynaptic profiles. Three-fourths of the postsynaptic processes could be identified as amacrine or ganglion cell. Of the 30 ribbons where both members of the dyad were identified, 15 were ganglion/amacrine cell pairs, 3 were ganglion/ganglion cell pairs, and 2 were amacrine/amacrine cell pairs. Amacrine cells received 37% of the contacts, about half of these postsynaptic amacrine cell profiles had reciprocal, feedback synapses, and the other half were nonreciprocally connected. Ganglion cell dendrites received 28% of the inputs. Almost all of the postsynaptic ganglion cell dendrites were dark, there being only one pale ganglion cell dendrite among the 25 ganglion/amacrine cell dyads.

CBBb. The soma of cell 13 was pear shaped, of medium diameter (4.5 μm), and located in the middle of the inner nuclear layer (Fig. 10). The cytoplasm was medium-dark. A single, thick primary dendrite extended toward the outer plexiform layer where it divided into six branches. These arborized at the bases of cone pedicles, but the contacts from cones could not be identified. The axon, like that of CBBc, was darker than the CBBb axon (Fig. 10) and paler than the rod bipolar cell axons. Upon entering the inner plexiform layer, the axon emitted two slender processes which could not be traced. One of these made a pair of synaptic contacts with processes in stratum 1 (Fig. 11). The axon dilated slightly at the junction of strata 2 and 3 and began to make many synapses within the neuropil. The axon twisted into a tangential orientation at the junction of strata 3 and 4, producing three branches that arborized in stratum 4. Most of the synaptic contacts were located in stratum 4 (65%) rather than in stratum 3 (35%) which was the reverse of the pattern exhibited by CBBc. Cell 13 received 46 chemical synaptic contacts, which were overwhelmingly (82%) from amacrine cell processes which did not themselves receive reciprocal contacts from cell 3. Reciprocally amacrine cell profiles provided the remaining inputs (20%). This bipolar cell made three small gap junction contacts with All processes. Cell 13 also provided 59 ribbon contacts to postsynaptic profiles, most of which (84%) were identifiable as amacrine or ganglion cells. Of the 44 ribbon synapses in which both members of the dyad were identified, 37 were ganglion/ amacrine cell pairs; 6 were ganglion/ganglion cell pairs, and 1 was an amacrine/amacrine cell pair. Amacrine cell profiles, one-third of which were reciprocally connected to the bipolar cell axon terminal and two-thirds of which were not, received 37% of the ribbon contacts. Ganglion cells received 47% of the
CBb

ribbon contacts. Two-thirds of these postsynaptic ganglion cell dendrites were pale and the rest were dark. Thus, in contrast to CBb, 24 of the 37 ganglion/mameric cell dyads had a pale ganglion cell dendrite as half of the dyad pair.

CBb and CBb had some features in common, but their differences were most striking. The axons had similar cytoplasmic density (darker than those of CBb), paler than those of Rb), they arborized in strata 3 and 4, and the somas were both adjacent to the outer plexiform layer. However, compared to CBb, the CBb soma was larger, and its axon made more
contacts in stratum 4 and fewer in stratum 3; it also received more inputs from nonreciprocally amacrine cell profiles and made more outputs to pale ganglion cell dendrites.

Core bipolar cells arborizing in sublamina c

Six core bipolar cells with axons arborizing in sublamina c were partially reconstructed. These proved to be of four types, which we have called CbAc, CbB, CbD, and CbE.

CbAc. There were two examples of this type, of which cell 1 was more complete. The soma of cell 1 was oval (8.8 μm wide, 11.9 μm high) with pale cytoplasm and was located in the middle of the inner nuclear layer (Table IV). Its single dendrite split into several branches in the outer plexiform layer where one received an interplexiform cell contact (Fig. 12). Another branch arborized near three cones, receiving from one at least three separate “flat” synaptic terminals (Boycott and Kolb, 1972) (Fig. 13). The axon branched immediately upon entering the inner plexiform layer, forming processes which were smooth in contour and pale (Fig. 14). Within strata 1 and 2, it arborized in a bushy knot of processes and made synaptic contacts.

Cell 1 received 28 chemical synaptic contacts from amacrine cells of several types (Table VI). Lobular appendages of the All amacrine cell provided a great proportion of the contacts (43%). Seven of 12 such inputs were rectified by a single All amacrine cell. Of the remaining 57% of the presynaptic amacrine contacts, only 1% received a reciprocal ribbon synapse back from cell 1. Cell 1 provided no ribbon contacts to postsynaptic processes, three-fourths of which could be identified as either other amacrine or ganglion cell dendrites. The remainder, which included 10 inputs of 39 (42%), were contact pairs, as no inputs from All were identified as such. A small number of inputs (5) were likely to be from a single All amacrine cell, as in this section. Most of the remaining postsynaptic amacrine cell profiles were nonreciprocating terminals, with the small remainder (10%) of the total making reciprocal connections with cell 1. Forty-one percent of the outputs from cell 1 were to ganglion cell dendrites, two-thirds of which were dark.

The axon of cell 4 resembled that of cell 3 in a number of respects but were different in some, missing some contacts about its correspondence to cell 1. The two cells were similar in having pale cytoplasm and a smooth axonal contour. The synaptic patterns of cell 4 were similar to that of cell 1 in that most of the inputs were from innerplexiform amacrine and All cells, and few were from reciprocal amacrine cells. The processes postsynaptic to cell 14 were similar to that of cell 1 in that most of the inputs were from innerplexiform amacrine and All cells, and few were from reciprocal amacrine cells. The processes postsynaptic to cell 14 were similar to that of cell 1 in that most of the inputs were from innerplexiform amacrine and All cells, and few were from reciprocal amacrine cells.
Figure 8. CBb$_2$ and CBb$_3$ reconstructions. Both are thinner than CBb in S3. CBb$_2$ arborizes primarily in S3; CBb$_3$ arborizes primarily in S4. a, input; b, output; §, gap junction with all cell; ©, interplexiform cell input. INL, inner nuclear layer; IPL, inner plexiform layer; OPL, outer plexiform layer.
report of a cone bipolar cell arborizing in sublamina a that receives ribbon-related contacts from cotes.

The axon branched upon entering the inner plexiform layer (Fig. 15), forming a rather bushy arborization. The dark axonal processes were varicose with swellings (2 to 3 μm in diameter), which were scalloped and angular in contour, interconnected by thin (0.3 to 0.6 μm) processes (Fig. 17). The axon ramified through strata 1 and 2 and the top part of stratum 3. Since its lateral extent exceeded the region photographed for the series, the arborization must have been wider than it appears in Figure 15. Most of its synaptic contacts were made within stratum 1.

Cell 15 received 38 chemically synaptic contacts from amacrine cells. Most (80%) were from nonreciprocal amacrine cell profiles, the rest (20%) being from reciprocal amacrine cells which received feedback ribbon contacts from cell 15. All amacrine cells provided no contacts to this cell. A total of 47 ribbon contacts were provided to postsynaptic profiles, most of which (70%) were identifiable as amacrine or ganglion cell. Of 20 fully characterized dyads, 13 were ganglion/amacrine cell pairs, and 7 were amacrine/amacrine cell pairs. Amacrine cells received 33% of all the ribbon contacts. More than half of the postsynaptic amacrine cells were nonreciprocal; about one-third were reciprocally connected to the bipolar cell; and very few were all cells. Ganglion cells received most of the ribbon contacts (46%), and just over half of these were dark.

Cell 5 assembled cell 15 in soma, dendritic and axonal diameters, and cytoplasmic density. The fine morphology of their axons was also similar in that each consisted of scalloped varicosities connected by thin necks. There were also basic similarities in synaptic pattern, such as having much input from nonreciprocal amacrine cells, less from reciprocal amacrine cells, and hardly any from all amacrines. Of the 25 dyads characterized for cell 5, 24 were ganglion/amacrine cell pairs (4 being All), and I was a ganglion/ganglion cell pair. The two cells appeared to differ in their relative numbers of outputs to amacrine and ganglion cells, evident, for example, in the smaller number of contacts between cell 5 and pale ganglion cells compared with cell 15. This discrepancy can probably be attributed to the higher percentage of unidentified postsynaptic processes for cell 15, since a certain portion of those presumably belong to slender, pale ganglion cell dendrites which lacked prominent clusters of ribbons, and further identification would thus bring the numbers into better agreement. The exact proportions of presynaptic reciprocal and nonreciprocal amacrine cell processes also differed, but this could be due to the incompleteness of the reconstruction for cell 15 (Fig. 15). Another difference between the pair was that cell 5 only arborized within strata 1 and 3, whereas cell 15 reached the top part of stratum 3. However, both processes of cell 15 that reached stratum 3 contacted the plexiform branch of an off cell, most of whose input from cell 15 was within strata 1 and 2. Perhaps the "arborization rule" for this axon type is flexible enough to be overridden when there is an opportunity for making particular synaptic connections.

**CB5**. The soma of cell 12 was pale with pale cytoplasm and located in the outer part of the inner nuclear layer (Fig. 18). The cell body emitted a single dendrite which divided immediately into at least five branches, one of which received an intercepting cell contact. The nature of the cone contacts on these dendrites could not be determined. A short axon emerged from the side of the soma and penetrated 4 to 5 μm into the inner plexiform layer before branching laterally (Fig. 19). The arborization was not bushy, but stratified parallel to the retinal layers, with one branch in the center of stratum 1 and the other in upper stratum 2. Individual axonal profiles were pale and smooth in contour (Fig. 19) and, therefore, impossible to distinguish in single sections from cross-sections of CB5 axons. Synaptic contacts between this axon and other neuronal profiles occurred about equally in strata 1 and 2.

The axis of cell 12 received 34 chemical synaptic contacts from amacrine cell processes. Amacrine cells not reciprocally

**TABLE IV**

<table>
<thead>
<tr>
<th>Morphological characteristics of sublamina a bipolar cells*</th>
<th>CB5*</th>
<th>CB5*</th>
<th>CB5*</th>
<th>CB5*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Soma diameter (μm)</td>
<td>8.8</td>
<td>8.8</td>
<td>8.8</td>
<td>8.6</td>
</tr>
<tr>
<td>Axon position (μm from IPL)*</td>
<td>13</td>
<td>14</td>
<td>9</td>
<td>16</td>
</tr>
<tr>
<td>Primary dendritic diameter</td>
<td>1.1</td>
<td>2.2</td>
<td>2.6</td>
<td>1.7</td>
</tr>
<tr>
<td>Primary axon diameter</td>
<td>1.4</td>
<td>1.1</td>
<td>0.8</td>
<td>1.7</td>
</tr>
<tr>
<td>Axon contour</td>
<td>Smooth</td>
<td>Variance &amp; Scalloped</td>
<td>Smooth</td>
<td>Smooth &amp; Delicate*</td>
</tr>
<tr>
<td>Axon stratification</td>
<td>S1, 2, 3</td>
<td>S1, 2</td>
<td>S1, 2, 3</td>
<td>S1, 2</td>
</tr>
<tr>
<td>Cyttoplasm density</td>
<td>Pale</td>
<td>S1, 2</td>
<td>Dark</td>
<td>S2</td>
</tr>
</tbody>
</table>

* See Table I legend for measurement criteria.

1 Cell type.
2 Cell number.
3 Measurement unavailable due to incomplete reconstruction.
4 IPL, inner plexiform layer.
connected to the bipolar cell provided the bulk of the contacts (83%), while reciprocal amacrine cells comprised the rest. There were no inputs from All cells. The axon of cell 12 made 47 ribbon contacts onto postsynaptic processes, 79% of which were identified as amacrine or ganglion cell. Amacrine cells received rather few ribbon contacts (18% of the total): most of these were to amacrine cell profiles which did not themselves receive ribbon contacts from cell 12, with a few to reciprocal and All amacrine cell processes. Ganglion cell dendrites, divided equally between dark and pale varieties, received the most contacts (61%). Of 27 dyads where both members were identified, 17 were ganglion/amacrine cell pairs and 10 were ganglion/ganglion cell pairs.

CBa1. The axon of cell 2 was broader than it was high. It contained pale cytoplasm (Fig. 5) and was located low in the inner nuclear layer (Fig. 14). The primary dendrite ascended without branching halfway to the outer plexiform layer before extending beyond the plane of the series. The axon was delicate and more difficult to trace than those of the other sublamina a cone bipolar cells, so that its arborization within the series is
patterns of connection from bipolar cells to ganglion cells

The axons of all three types of bipolar cells in sublamina b contacted the dendrites of 65% ganglion cells, and both CbB and CbC, but not CbA, contacted large, pale dendrites probably belonging to on-off cells. The axons of three of the four types of bipolar in sublamina a (CbB, CbC, and CbB) contacted the dendrites of off-ON cells and pale dendrites probably belonging to off cells.

Individual cone bipolar cells commonly directed many ribbon contacts to a single ganglion cell. The greatest number of contacts on a ganglion cell from a single cone bipolar was 47 (CbB cell in Fig. 20a). There were examples of diverging connections, too, in that individual cone bipolar cells sometimes contacted several morphologically different kinds of ganglion cells. In sublamina b, for example, CbB cells (cell 15) contacted an off-ON cell (Fig. 20b) 23 times and a large, pale dendrite (Fig. 20b) 7 times. From the ganglion cell’s point of view, two kinds of divergence were observed. A single ganglion cell received contacts from two cone bipolar cells of the same type. Thus, one on-ON cell (Fig. 20a) received 69 CbB contacts, 47 from cell 1 and 22 from cell 3. A ganglion cell also received contacts from two or more cone bipolar cells of different type. Thus, an on-ON cell (Fig. 20e) received 53 contacts from CbB, and CbB, and CbC. Other examples of these patterns of bipolar-to-ganglion cell contact in both sublaminae are illustrated in Figure 28.

Discussion

Definition of bipolar cell types. We have reconstructed 15 bipolar neurons from serial electron micrographs in order to establish a scheme for their classification. The reconstruction approach was favorable for this purpose because 1. A preserved information simultaneously about external morphology, cytoplasmic features, and synaptic circuitry. This approach also allowed us to compare neurons within a tiny region of a single retina, which means that differences observed between cells could not be attributed to differences in retinal eccentricity (Ansell and Wässle, 1979) or interindividual variation. A “type” was defined when a cell exhibited a distinctive set of associations between morphology, cytotax, and synaptic patterns. In all, there were evidence for one type 60 bipolar and seven type of cone bipolar cells. We believe these represent fundamental cell types in somewhat the same sense as axotopole are “identified” in invertebrates.

Multiple examples were obtained for two types, the rod bipolar and the cone bipolar, CbB. All of the examples of each type were strikingly similar. Thus, the three rod bipolar cell axons were almost identical in form, diameter, and the depth in the inner plexiform layer where they dilated and began to make synapses. These axons were remarkably consistent in their absolute numbers of synaptic inputs (25, 39, 43) and outputs (27, 28, 33) and also in their ratios of specific types of presynaptic and postsynaptic processes (Tables II and III). The four reconstructed CbB axons also resembled each other remarkably in morphology, diameter, and the depth in the inner plexiform layer at which they formed synapses. Their arborizations, which were much more elaborate than those of the rod bipolar cell, could not be reconstructed in their entirety. Consequently, the absolute numbers of their inputs and outputs could not be determined; however, the ratios of specific types of contacts were remarkably consistent among the four axons. For example, inputs from reciprocal axons of CbB cells were 24, 25, 28, and 30% of all inputs (Table II); similarly, the outputs to amacrine cells were from 29% to 33% and outputs to ganglion cells were 19% to 30% of all outputs (Table III). Wässle and colleagues (Wässle and Nieman, 1978; Wässle et al., 1978, 1985a, b) have reported in light microscopic studies of
<table>
<thead>
<tr>
<th>Source of inputs</th>
<th>CBa</th>
<th>CBb</th>
<th>CBc</th>
<th>CBd</th>
<th>CBa</th>
<th>CBb</th>
<th>CBc</th>
<th>CBd</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
<td>12</td>
<td>5</td>
<td>2</td>
<td>1</td>
<td>12</td>
<td>5</td>
<td>2</td>
</tr>
<tr>
<td>All amacrine</td>
<td>43%</td>
<td>29%</td>
<td>0%</td>
<td>6%</td>
<td>0%</td>
<td>12%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Reciprocal</td>
<td>7%</td>
<td>7%</td>
<td>18%</td>
<td>37%</td>
<td>15%</td>
<td>59%</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nonreciprocal</td>
<td>50%</td>
<td>64%</td>
<td>87%</td>
<td>57%</td>
<td>83%</td>
<td>29%</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Total no. of inputs

100% 100% 100% 100% 100%

*Conventions for quantifying inputs are as in Table II.

**Cell type.

***Cell number.

### TABLE VI

<table>
<thead>
<tr>
<th>Conventions for quantifying outputs are as in Table III. Post synaptic densities on all cells were never quantified for other cells, even though the All profile was prominently positioned at the dendrite.</th>
</tr>
</thead>
</table>

<table>
<thead>
<tr>
<th>CBa</th>
<th>CBb</th>
<th>CBc</th>
<th>CBd</th>
<th>CBa</th>
<th>CBb</th>
<th>CBc</th>
<th>CBd</th>
<th>CBa</th>
<th>CBb</th>
<th>CBc</th>
<th>CBd</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>1</td>
<td>1</td>
<td>2</td>
<td></td>
<td>1</td>
<td>1</td>
<td>2</td>
<td></td>
<td>1</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>To amacrine cells</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>All amacrine</td>
<td>18%</td>
<td>29%</td>
<td>0%</td>
<td>6%</td>
<td>18%</td>
<td>29%</td>
<td>0%</td>
<td>6%</td>
<td>18%</td>
<td>29%</td>
<td>0%</td>
</tr>
<tr>
<td>Reciprocal amacrine</td>
<td>1%</td>
<td>2%</td>
<td>10%</td>
<td>21%</td>
<td>5%</td>
<td>23%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Nonreciprocal amacrine</td>
<td>15%</td>
<td>8%</td>
<td>19%</td>
<td>21%</td>
<td>10%</td>
<td>18%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total amacrine</td>
<td>24%</td>
<td>39%</td>
<td>22%</td>
<td>45%</td>
<td>18%</td>
<td>41%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>To ganglion cell</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Dark ganglion cell</td>
<td>28%</td>
<td>16%</td>
<td>21%</td>
<td>52%</td>
<td>27%</td>
<td>36%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Pale ganglion cell</td>
<td>13%</td>
<td>21%</td>
<td>19%</td>
<td>24%</td>
<td>24%</td>
<td>36%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total ganglion cell</td>
<td>41%</td>
<td>39%</td>
<td>46%</td>
<td>50%</td>
<td>21%</td>
<td>44%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Undifferentiated</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>25%</td>
<td>22%</td>
<td>21%</td>
<td>34%</td>
<td>21%</td>
<td>45%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>100%</td>
<td>100%</td>
<td>100%</td>
<td>100%</td>
<td>100%</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total no. of outputs</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>69</td>
<td>50</td>
<td>47</td>
<td>47</td>
<td>22</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

**Cell type.

***Cell number.

horizontal and ganglion cells that cells of a particular type at a given locus in one retina are remarkably constant in soma size and dendritic field and that each type forms a regular array in the retina mosaic. This appears to be true also for particular types of bipolar cell. Thus, CBa axons form a highly regular array with a nearest neighbor distance of 10 ± 1 μm (mean ± SD; Freed and Sterling, 1983). One feature that appeared to be constant was the exact depth of the soma in the inner nuclear layer of RB cells (Fig. 6). Conservation of form and synaptic connection but not of exact soma position is a hallmark of the neurons in vertebrate nervous systems that have been termed "identified" (Sterling, 1982).

Most of the types we have defined by serial reconstruction correspond to cells identified in previous studies, either by Golgi impregnation (Famiglietti, 1981; Kolb et al., 1981) or by intracellular recording and horseradish peroxidase injection (Kolb and Nelson, 1983; Nelson and Kolb, 1983). We review the correspondences in detail below for two reasons. First, they strengthen the definition of each cell type, especially where only one example was seen, and they establish that, for at least one small part of the mammalian nervous system, there is a correlation at the neuron level between form and function. Second, by knowing both the anatomical circuitry and, for some, the electrophysiology, it is possible to formulate specific hypotheses regarding why there might be such a diversity of cell types.

Correspondence between morphology, synaptic circuitry, and physiology. The cell identified as a rod bipolar corresponds precisely in morphology and cytoLOGY to the rod bipolar (RB) identified by previous authors (Boycott and Kolb, 1972; Kolb, 1979; Famiglietti, 1981; Kolb et al., 1981). The rod bipolar cell dendrite receives ribbon-related input from rods (e.g., Kolb, 1979). The reciprocal amacrine cell profiles, which provided 80% of the input to its axon terminal, are believed to derive from GABA-accumulating amacrine cells A13 and A17 (Kolb et al., 1981; Freed and Sterling, 1982; Pourcho and Goebel, 1980). The rod bipolar cell is hyperpolarized to light (Nelson and Kolb, 1983) and is thought to be inhibitory because its hyperpolarization apparently causes depolarization of the All amacrine to which it provides major input (Famiglietti and Kolb, 1975, Nelson, 1982).
Figure 15. CBa2 reconstructions. Above, Cell 5 rotated 30° and cell 15 rotated 180° about the Y axis. Below, Both cells rotated 90° about the X axis. Axon varicosities are prominent in all cases. O: output; A: input; +, AII inputs; @, on-centerbursting cell input. INL, inner nuclear layer; IPL, inner plexiform layer; OPL, outer plexiform layer.

from the depolarizing AII amacrine to the on ganglion cells (Figs. 8 and 20; see also Nelson and Koli, 1983; Sterling, 1983).

The type of cone bipolar cell we call "CBa2," with an axon narrowly stratified in stratum 3 and upper stratum 4, corre-
sponds to the wide-field "cb" of Koli et al. (1981) and probably to Fiumaguetti's (1980) "cbp." Although this was illustrated by Koli et al. (1981) as branching only within stratum 3, that study divided the inner plexiform layer into equal fifths (for
The type of cone bipolar cell we call "CBA", with its smooth axon branching tangentially in strata 1 and 2, resembles the "smooth, flat" cone bipolar described by Boycott and Kolb (1973). It does not correspond clearly, however, to any type in the recent schemes of Kolb et al. (1981) or Famiglietti (1981) because CBA ramifies in both strata 1 and 2 whereas, ch2, the only possible candidate in the scheme of Kolb et al. (1981), is described as restricted to the border between these strata. The response of CBA to light has not yet been determined.

The type of cone bipolar cell we call "CBA", with a thin axon ramifying solely above the border between sillonina and a,b, corresponds to type "ch2" of Kolb et al. (1981) and to type "nch2" of Famiglietti (1981). It has been shown to receive excitatory inputs from cones (Nelson and Kolb, 1983), but its responses to light have not been determined.

From these correspondences several points emerge. First, the categories of bipolar cell identified in the retina of one cat by reconstruction are also found in the retina of other cats, which is part of the evidence that they are indeed specific cell types. Second, the size of the patch within which all of these types were found ranged from 0.06' x 0.0' on the retina. This is less than the area covered by an octagonal cell dendritic field and is in effect a "point" in the visual field. It follows that the visual image at every point on the cat retina must be analyzed by one type of rod bipolar cell and 8 to 10 types of cone bipolar cells, very much in the sense first suggested by Maturana et al. (1960). This is somewhat startling, since the cell is specialized for night vision and generally is considered rod dominated. One wonders what might be the function of so many cone bipolar types.

Possible functions for multiple types of cone bipolar cells. One function for cone bipolar cells might be to convey, separately or in specific combinations, cone signals of different spectral sensitivities. The only type of cone bipolar which has direct recordings has a peak sensitivity at 556 nm (Nelson, 1977), but behavioral studies and recordings from retina and lateral geniculate nucleus (Daw and Perlmutter, 1969; Farbman and Daw, 1970; Cleland and Lavick, 1974) provide evidence for a second cone type with a peak sensitivity at 450 nm. Recent ganglion cell recordings confirm these observations and provide evidence for a third type with peak sensitivity at 500 nm (Ringo and Weller, 1961; Zrenner and Weinrich, 1961). If information from three cone types regarding color is distributed to both the
on and the off sublaminae, it would not be surprising if many distinct types of cone bipolar cells were required. This would be especially true if, as is common, the information were conveyed in chromatically and spatially antagonistic forms (Daw, 1973). It is unknown whether the cone bipolar types described here are color coded.

Another important set of functions for the cone bipolar neurons is, oddly enough, to convey rod signals to ganglion cells via multiple pathways. Koff (1979) reported that the rod bipolar cell provides no direct contacts to ganglion cells, although they had been seen in the rabbit retina (Flavell and Yarnska, 1967). Our reconstructions reveal a few such contacts (Fig. 7, Table III; see also Freed and Sterling, 1980), but we agree that there is no evidence for a major direct pathway. The rod bipolar cell apparently has access to ganglion cells mainly via its multiple connections with the AII amacrine cell which in turn contacts at least six types of cone bipolar. The AII makes gap junction contacts in sublamin b with CBb and, to a lesser extent, with CBs and CBb. The AII bipolars apppendages make chemical contacts in sublamin a with CBa and, to a lesser extent, with CBa and CBb. The rod signal is also transmitted laterally in the outer plexiform layer from rods to cones, ap-
Figure 7B. Electron micrograph of Cbα, axon. This type resembles Cbα, in some respects because its axon is pale and smooth in contour. Arrow, amacrine input; circle, synaptic ribbon; AM, amacrine cell axon.

Figure 8B. Schematic drawing of seven ganglion cells and three isolated dendrites that received multiple bipolar inputs. Cells A, D, and E in β cells, B and G, off-β cells; C, possible cell; F, small ganglion cell. Dendrites: H, possible on-α cell; J, possible off-α cell dendrite; R, dendrite L, possibly on-α, although it is less explicit in contour than H of J, received both cone and rod bipolar inputs. See the text for details. GCL, ganglion cell layer; INL, inner nuclear layer; IPL, inner plexiform layer.


Famiglietti, V. R., Jr., and H. Kohl (1975) A bistriated amacrine cell and synaptic circuitry in the inner plexiform layer of the retina. Brain Res. 102: 293-300.


