CHAPTER 2

Anatomy of vision

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Introduction

The neural infrastructure of visual processing is spread widely throughout the central nervous system. There are over 30 separate cortical areas implicated as substrates for the various functional attributes of vision but the majority of the visual input to these areas passes initially from the retina via the lateral geniculate nucleus to the striate cortex.

The aim of this chapter is to provide an overview of the structural organization of these initial elements of the visual pathways that provide the raw visual data for the various widespread cortical divisions. In addition, the aim is to provide an introduction to other subcortical pathways involved in visual processes as well as a basis for comprehending the consequences for visual field representation of damage to the initial parts of the visual pathway (see Ch. 20).

Retina

The retina is the eye's innermost coat, lining the posterior two-thirds of the internal surface. It is a thin, mostly transparent layer of tissue that extends forward from the optic disc to its anterior limit, the ora serrata, where it is continuous with the epithelial layers of the ciliary body.

The visual system's photosensitive components are located in the retina. However, the retina represents more than a mere phototransduction apparatus, as its origin as an outpouching of the brain belies. Its cytoarchitecture underlies sophisticated post-receptor processing of the response of the photosensitive cells (for comprehensive reviews see Masland 2001; Sterling & Demb 2004; Wässle 2004).

The retina originates as two primordial layers of invaginating epithelium. An outer layer, identified as the retinal pigment epithelium, and an inner layer that ultimately becomes the multilaminar neural retina. This layered organization of the retina and its nomenclature are described in **Figure 2.1A**.

The histological appearance of the retinal layers shows the distribution of the various cell bodies and processes but fails to reveal their organization as a number of morphologically and physiologically distinct cell types that are organized into vertical (through the layers) and lateral (within layers) paths of cellular interactions. The outermost laminae, adjacent to the pigment layer (retinal pigment epithelium, RPE), are where the majority of photosensitive cells are located. These photoreceptors, the rods and cones, represent the site where the transduction of light energy into the bioelectrical activity essential for vision is accomplished. Within the vertical path through the retina, information flow passes predominantly from the photoreceptors via the second-order neurons, the bipolar cells, to the third-order neurons, the ganglion cells, and thence from the retina to cellular targets centrally in the brain. At successive stages of this vertical path, cellular responses are influenced by lateral interactions, firstly with horizontal cells at the level of the photoreceptors and later by amacrine cells in the inner retina at the level of the ganglion cells. These separate paths are indicated schematically in **Figure 2.1A**.

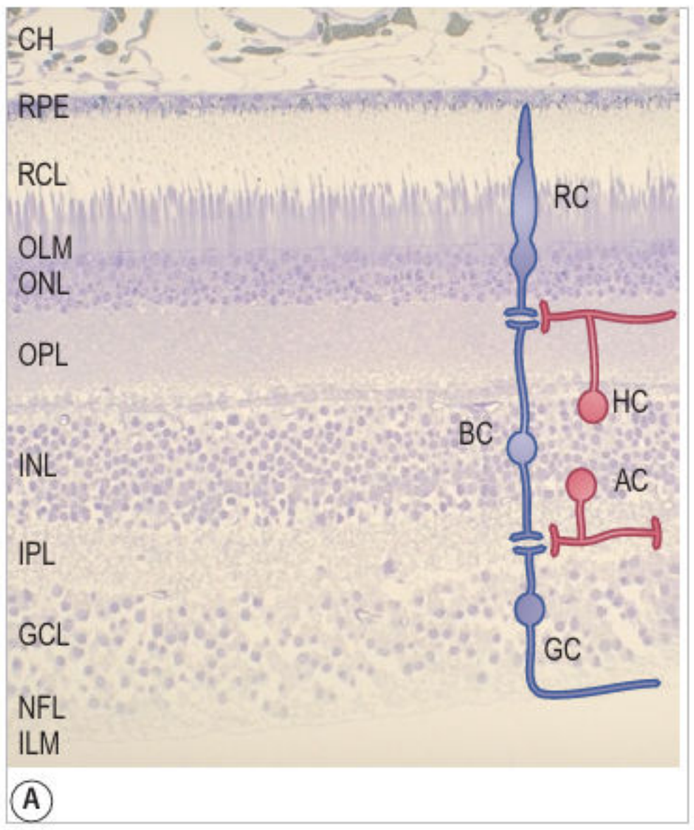

Figure 2.1A Transverse section through the mid-peripheral retina and choroid (CH) of a macaque retina stained with toluidine blue. Scale bar: 50 μm. The labels on the left side indicate the conventional nomenclature for the individual layers. RPE, retinal pigment epithelium; RCL, rod and cone layer; OLM, outer limiting membrane; ONL, outer nuclear layer; OPL, outer plexiform layer; INL, inner nuclear layer; IPL, inner plexiform layer; GCL, ganglion cell layer; NFL, nerve fibre layer; ILM, inner limiting membrane. The right side of the section is overlaid with diagrammatic representations of the vertical (blue) and horizontal (red) paths of cellular interaction within the retina. RC, receptor cell; BC, bipolar cell; GC, ganglion cell; HC, horizontal cell; AC, amacrine cell.

Foveal specialization

Prior to considering the cellular relationships in the retina, it is important to emphasize that its organization is not homogeneous. At the centre of the retina on the visual axis is a region of approximately 5 mm in diameter known as the macula lutea (yellow spot), so called as a consequence of the xanthophyll derivates found within the retinal cells of the region, which impart a yellowish hue. At the centre of this macular region is a depression or pit, the fovea, with a diameter of approximately 1.5 mm. Peripheral to the fovea are the parafoveal and perifoveal regions, while at the foveal centre is the foveola.

The cellular organization is adapted in the fovea to allow the finest level of visual resolution in the roughly central 5 degrees of visual angle. Specific mention will be made of the consequences of the adaptation of the central retina for the organization of the visual pathways in the relevant sections below. Microscopically, however, the most obvious modification of this retinal region is the reduced thickness (**Fig. 2.1B**). Consequently, the path of light to the photoreceptors is comparatively unobstructed by overlying tissue, a factor that is further enhanced by the absence of the vasculature from the foveal centre.

Pigment epithelium

This, the outermost layer of the retina, is a single-cell-thick layer of tessellating, polygonal and relatively cuboidal epithelium (**Fig. 2.1A**: RPE; **Fig. 2.2A**). It is continuous rostrally with the outer, pigmented epithelial layer of the ciliary body. The basal surface of these cells provides the inner basal lamina component of Bruch's membrane, which separates the pigment epithelium from the adjacent choriocapillaris, the vascular supply that provides for the outer retina including the photoreceptors. Gap junctions, anchoring

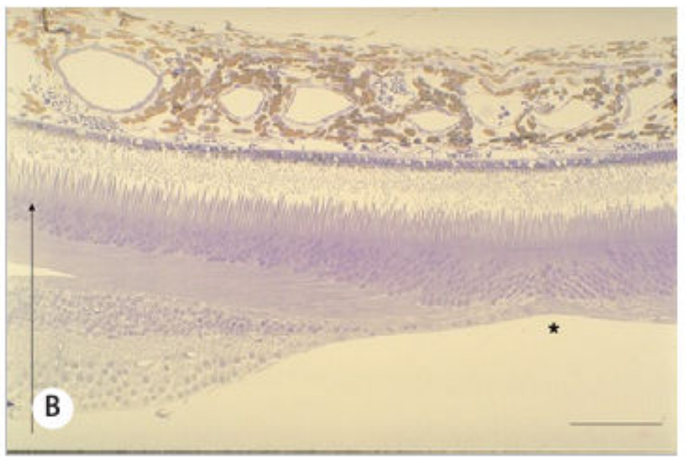

Figure 2.1B Transverse section through the foveal region stained with toluidine blue. Scale bar: 50 μm. The centre of the fovea, the foveola, is indicated by the asterisk towards the right-hand edge of the micrograph. The left-hand edge is the parafoveal margin and the layer designations defined in **Figure 2.1A** apply there. The foveola is characterized by the virtual absence of the retinal layers vitreal to the outer nuclear layer. The arrow indicates the direction of the light path towards the photoreceptors. Clearly, peripheral to the foveal region, light must pass through the layered array of cells and processes that comprise the retina. However, at the fovea, on which the image is normally fixated, the obstruction is minimal.

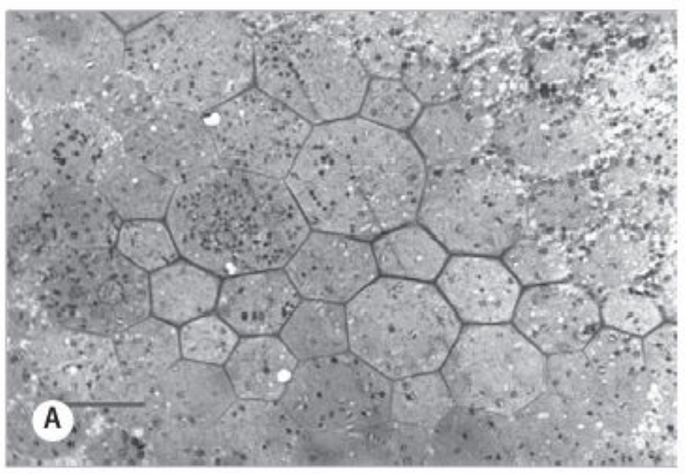

Figure 2.2A A tangential section through human pigment epithelial cells. Scale bar: 25 μm. The tight junctions between the cells enhance the outline of the cells. There is a large size and shape difference, varying from pentagonal to octagonal. The specimen was taken from a 70-year-old; in younger individuals, there is less variation, with the hexagonal form dominating.

junctions and occluding junctions are found between pigment epithelial cells, the latter towards the apical surface and forming a part of the blood–retina barrier. The apical surface of each of these cells is intimately associated with the rods and cones (**Fig. 2.2B**), and this relationship, which can be disrupted by retinal detachment, is critical for sustaining the photosensitive properties of the rods and cones. The pigment within the cells of this layer is melanin, which is packaged in melanosomes distributed mainly towards the apical (inner) surface of the cells. Melanin will absorb light that passes through the retina and therefore acts to prevent light scatter, thereby reducing image degradation.

Photoreceptor cells

Retinal distribution

Rod photoreceptors are adapted to perform best in scotopic conditions whereas cones are specialized for brighter, photopic, circumstances. They contain different light-sensitive photopigments. There is one basic rod type but cone photoreceptors comprise three major types that differ with respect to their wavelength specificities and are thus subdivided into red, green and blue cone types, also known respectively as L (long-wavelength sensitive), M (mid-wavelength sensitive) and S (short-wavelength sensitive) cones.

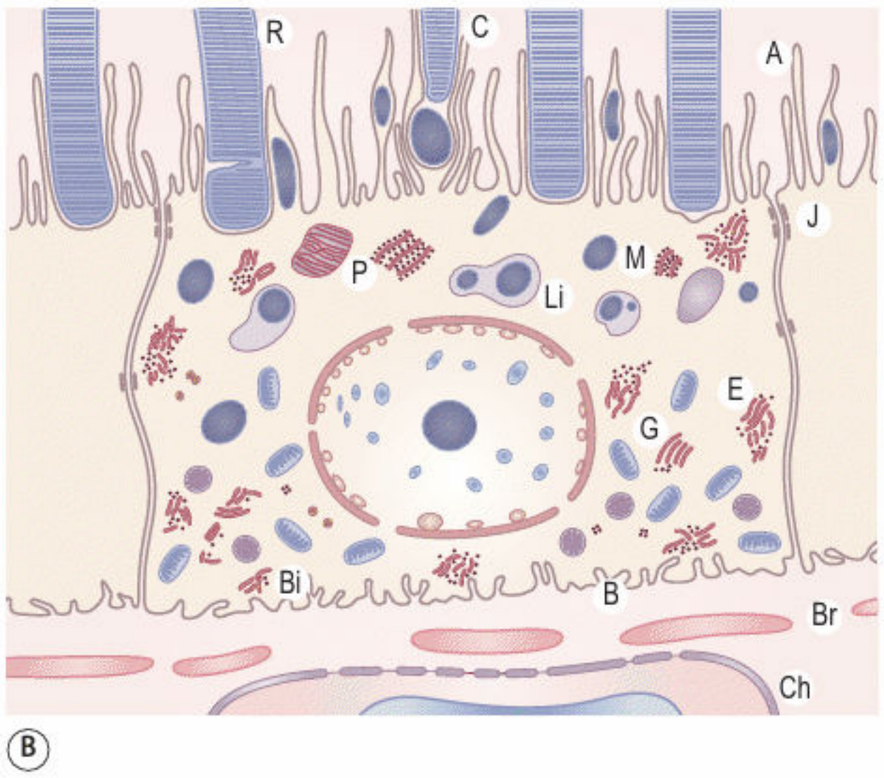

Figure 2.2B Schematic diagram of a retinal pigment epithelial cell. A, apical processes; B, basement membrane; Bi, basal infoldings; E, endoplasmic reticulum; G, Golgi complex; J, tight junction; Li, lipofuscin granule; M, melanosome; P, phagosome; Br, Bruch's membrane; Ch, choriocapillaris; R and C, outer segment tips of rod and cone photoreceptors.

Rods comprise the largest proportion of receptors. Classical accounts describe rods amounting to 120 million/eye while cones total roughly 6 million/eye (Østerberg 1935). However, a recent study utilizing a more appropriate sampling procedure estimates the (mean) rod population at approximately 90 million and that for the cone population at about 4.5 million for each eye, although there appears to be a great deal of individual variability (Curcio et al 1990).

The far greater number of rods implies a much greater density of this photoreceptor but there is important regional specialization in photoreceptor density across the retina. Rods are present at higher density except in the macular region and at the extreme peripheral margin, where cones dominate (Williams 1991). Rods are virtually absent from the centre of the foveal pit (foveola) and are at their highest density at the perifoveal border, especially at its superior aspect (Curcio et al 1990).

The three cone types are not present in equal numbers. It has been estimated that there are roughly 2.9 million red cones, 1.4 green cones and 0.2 million blue cones (Oyster 1999). In addition to their unequal numbers, the different cone types are also distributed differently across the retina; the presence of only red and green cones at the foveola means that we are normally tritanopic in the central fovea (Curcio et al 1991).

Morphology

Both photoreceptor cell types have an outer segment, which is the photosensitive compartment of the cell, joined to an inner segment by a narrow stalk containing a modified cilium. For rods, an outer fibre extends from the inner segment to the cell body region whereas, in general, cone inner segments are immediately adjacent to the cell body. An inner fibre (or axon) projects from the cell body of both receptor types with terminal end feet including synaptic regions that are specialized for each cell type (**Fig. 2.3A**).

Outer segment

The outer segments of rods are of a relatively long, tubular form while those of cones, at least outside the foveal region,

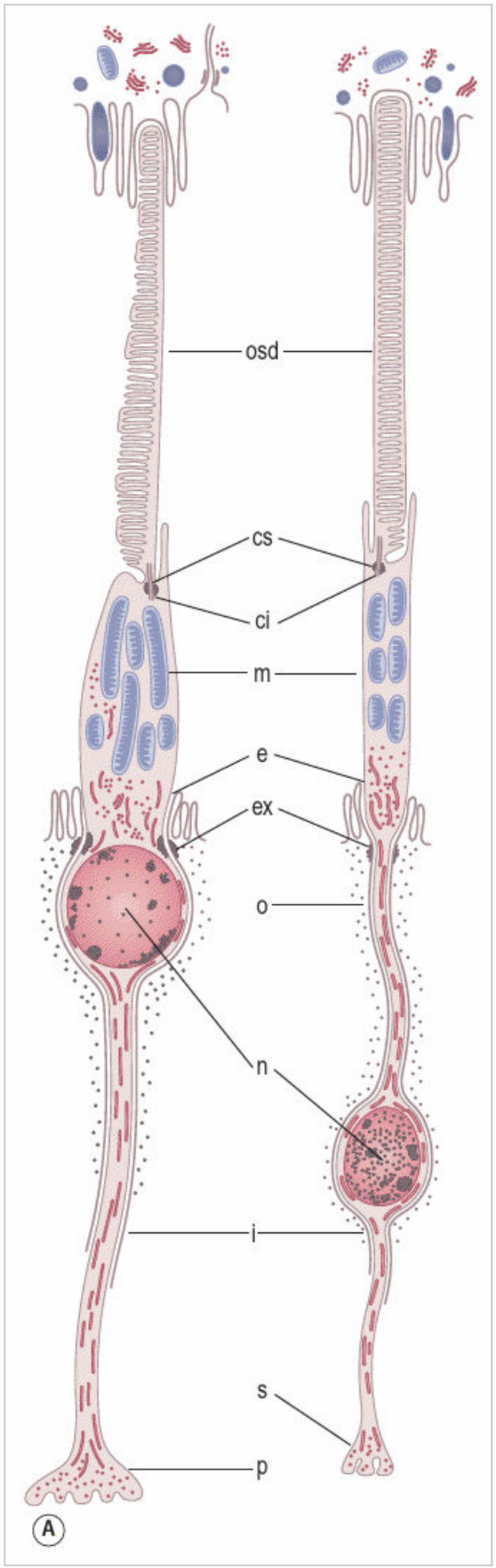

Figure 2.3A Schematic diagrams of a cone (*left*) and rod (*right*). osd, outer segment discs; cs, connecting stalk; ci, cilium; m, myoid; e, ellipsoid; ex, location of outer limiting membrane; o, outer fibre; n, nucleus; i, inner fibre; s, spherule; p, pedicle.

are shorter and conical. Within the fovea, the distinction is not so easily made between rods and cones, with both receptor types having similar cylindrical morphology in this region.

Each outer segment comprises a stack of membranous sacs or discs which incorporate the visual pigment molecules that are the basis of photosensitivity. In rods, these discs can be considered an intracellular organelle in that, except at the base of the outer segment where the discs are produced, they are distinct intracellular compartments separate from the cell membrane (**Fig. 2.3B**). In cones, by

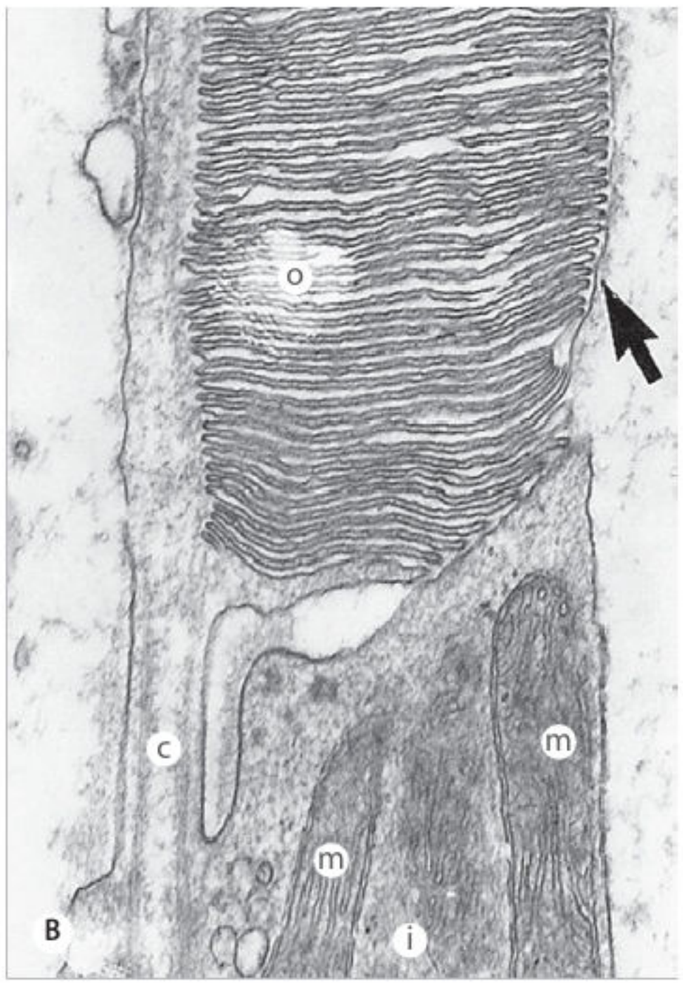

Figure 2.3B Rod photoreceptor of a rhesus monkey at the junction between inner and outer segments. i, inner segment; o, outer segment; c, cilium within connecting stalk; m, mitochondrion. A few discs adjacent to the cilium are continuous with the plasma membrane where they are formed but the remainder are detached from the plasma membrane and are free-floating within the outer segment (*arrow*).

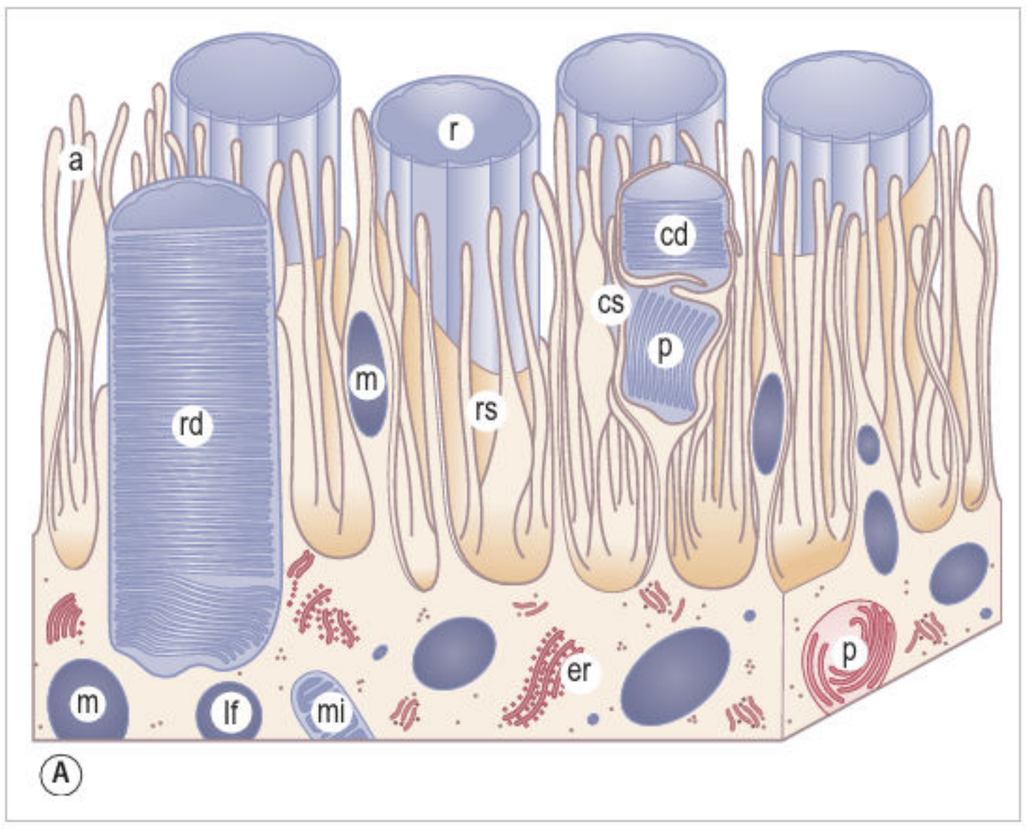

Figure 2.4A Schematic representation of the relationship between rods, cones and pigment epithelial cells. Most of the apical processes of the pigment epithelial cells are fine and finger-like while others form a sheath investment. a, apical process; cd, cone outer segment discs; cs, cone sheath; er, rough endoplasmic reticulum; gl, Golgi complex; lf, lipofuscin granule; m, melanosomes; m', mitochondrion; p, phagosomes – one of them about to enter the body of the pigment epithelial cell; r, rod; rd, rod outer segment discs; rs, rod sheath.

contrast, the discs are produced by a similar infolding of the plasma membrane at the base, but this organization is retained throughout the entire extent of the outer segment so that the disc's internal face is, in the main, part of the massively increased extracellular surface of the cell membrane.

The discs are not static or permanent. In macaque rods, they are removed or shed from the outer segment tips and renewed by continued production at the outer segment base (Young 1971). This cycle of disc shedding and renewal has been shown to be under circadian regulation in various non-primate species with the different photoreceptor types expressing distinct phases. In cone outer segments the discs are shed at the end of the day, while in rods the outer segment tips slough off during the transition from dark to light at the end of the night. The tips of the photoreceptor outer segments are enveloped by processes arising from the apical surfaces of the adjacent pigment epithelial cells (**Figs 2.2B** and **2.4A**). As the outer segment discs are shed, they are phagocytosed in groups of about 10 to 20, or more, by these cells (**Figs 2.4A** and **2.4B**) (Young & Bok 1969).

The extracellular region between the pigment epithelial cell processes and the photoreceptor outer segments, the so-called subretinal space, is occupied by the interphotoreceptor matrix. This is a mixture of proteins and glycosaminoglycans that may possess adhesive properties acting to preserve the close apposition of the adjacent cell layers (Chu & Grunwald 1990). In addition, at least one particular constituent, interstitial retinoid-binding protein, has an important role in transferring molecules essential in the phototransduction regeneration process, to and from the retinal pigment epithelial cells (Bok 1990).

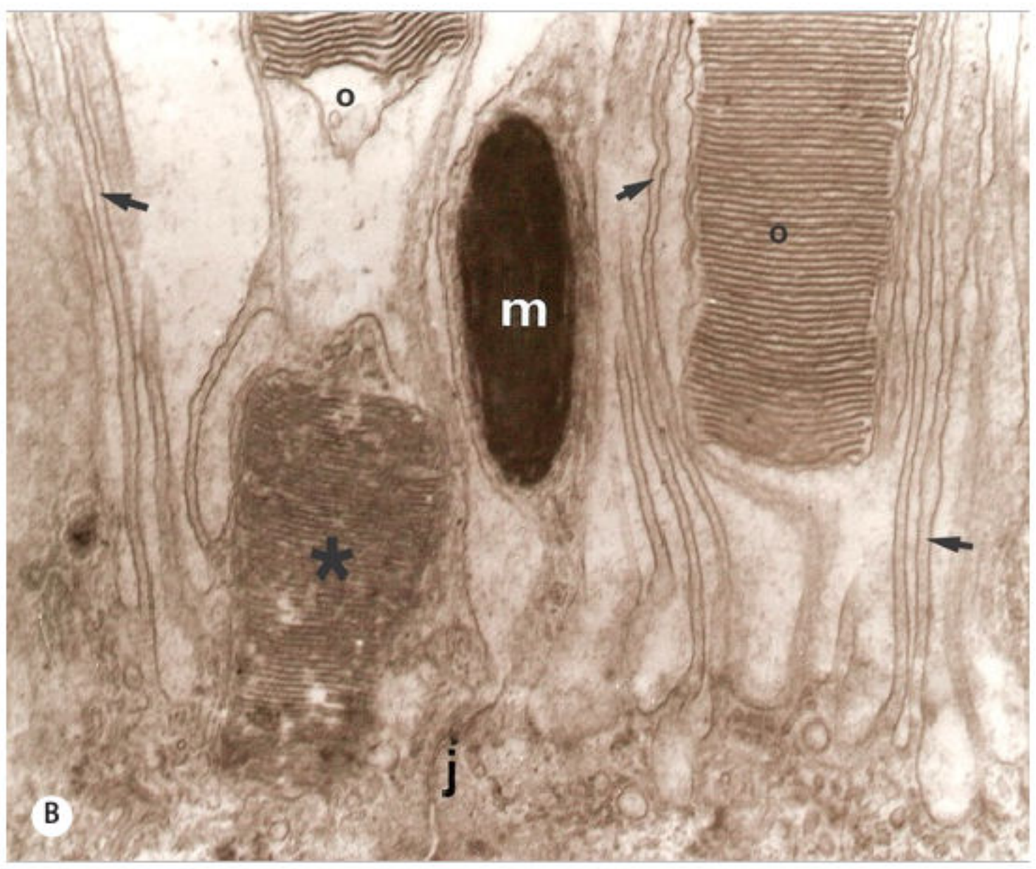

Figure 2.4B Electron micrograph of pigment epithelial apical processes and adjacent receptor outer segments of rhesus monkey. The apical processes, some of which are indicated by arrows, surround the tips of the outer segments. A group of approximately 50 outer segment discs from one of the receptor outer segments (*), in the process of sloughing off, has detached and is partly buried within the epithelial cell preliminary to the formation of a phagosome. m, melanosome; o, outer segment tips.

Inner segment

The inner segment is generally larger in cones than rods outside the foveal region but of roughly equal size in the fovea. It is subdivided into two regions: the region closer to the outer segment is known as the ellipsoid, the other as the myoid (see **Fig. 2.3A**). The ellipsoid houses a large concentration of mitochondria and thus represents the powerhouse

of the cell, while the myoid, occupied by endoplasmic reticulum and Golgi apparatus, is a region specialized for the protein production and packaging necessary for the renewal of discs at the base of the outer segment (Steinberg et al 1980) as well as the photopigment components embedded in those membranes (Bok 1985).

Just as the outer segments of photoreceptor cells are separated from each other by enveloping processes of the apical surface of the pigment epithelial cells, the inner segments of adjacent photoreceptors are similarly separated, but by processes of Müller cells. These are large glial, or supporting, cells that extend throughout the entire depth of the retina (**Fig. 2.5**). They form specialized tight junctions (maculae adherens) with the inner segments at their bases as well as with other Müller cell processes, forming a histologically defined retinal layer, the external (outer) limiting membrane, which is the inner border of the subretinal space (**Figs 2.1A** and **2.6**).

An outer fibre connects the inner segment to the cell body of the photoreceptor. This fibre is of almost negligible length in cones outside the foveal region, but in the foveola the cone cell body is displaced along the outer fibre. Outside the foveal region, the different lengths of their outer fibres result in the cell bodies of cones and rods, and the nuclei they include; being largely segregated in the outer nuclear layer, the cone cell bodies lying adjacent to the outer limiting membrane separate from several rows of rod nuclei (**Fig. 2.6**). An inner fibre, or axon, extends from the cell body region to terminate in a synaptic end foot. At the fovea, these axons belong predominantly to cones and are particularly evident as Henle's fibre layer (**Fig. 2.6**).

The axon terminal regions are morphologically specialized according to the photoreceptor cell type. Rod terminals, or spherules, are relatively spherical while the different cone types each have larger, flattened terminals known as pedicles (see **Figs 2.3A** and **2.6**). These can have small, laterally extending processes which are frequently in gap junction contact with other cone pedicles (Hornstein et al 2004) as well as with rod spherules (Haverkamp et al 2000).

The spherules and pedicles are located within the outer plexiform layer (see **Fig. 2.1A**). As found for their cell bodies in the outer nuclear layer, the different synaptic terminals are partitioned within this layer; rod spherules form several sublaminae towards the border with the outer nuclear layer, while the cone pedicles are distributed further towards the inner margin of the outer plexiform layer.

Invaginations within the photoreceptor terminal regions represent specialized regions for synaptic interaction with other retinal neurons. A flat plate, the 'synaptic ribbon', is found in the photoreceptor component of this specialized region, oriented orthogonally to an electron-dense region of presynaptic membrane and a synaptic ridge (**Fig. 2.7A**). Synaptic vesicles are associated with both sides of the ribbon and the function of the ribbon may be for rapid channelling of synaptic vesicles to their docking sites at the presynaptic membrane (Rao-Mirotznik et al 1995).

Rod spherules generally possess a single invagination (**Fig. 2.7A**) whereas there may be up to 50 invaginations found in the more complex and larger, flattened pedicle terminals (**Fig. 2.7B**) (Chun et al 1996). At these synaptic specializations, contacts are made with bipolar cell dendrites and the processes of horizontal cells. In each cone invagination, a central bipolar cell dendrite is flanked by two horizontal cell processes found deeper within the invagination, forming an arrangement known as a triad (**Fig. 2.7C**). The term triad implies only three components but the number of processes, especially within the invaginations of rod spherules, can exceed this number.

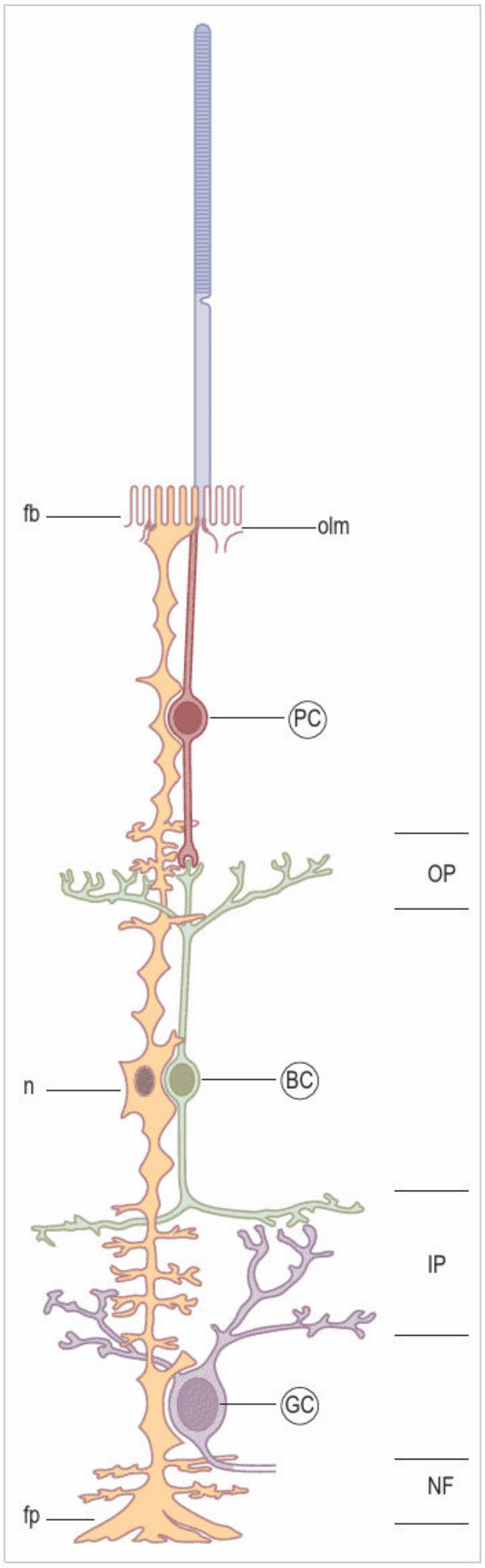

Figure 2.5 Schematic representation of a Müller cell. The soma (n) lies in the inner nuclear layer and its processes form numerous fine branches penetrating the neuropil of the two plexiform layers and the nerve fibre layer. Müller cells terminate in the outer retina in the form of microvilli-like fibre processes (fb) which penetrate the outer limiting membrane (olm) and surround the photoreceptor inner segments. At their inner retinal limit, Müller cells broaden to form footplates (fp) at the inner limiting membrane. OP, outer plexiform layer; IP, inner plexiform layer; NF, nerve fibre layer

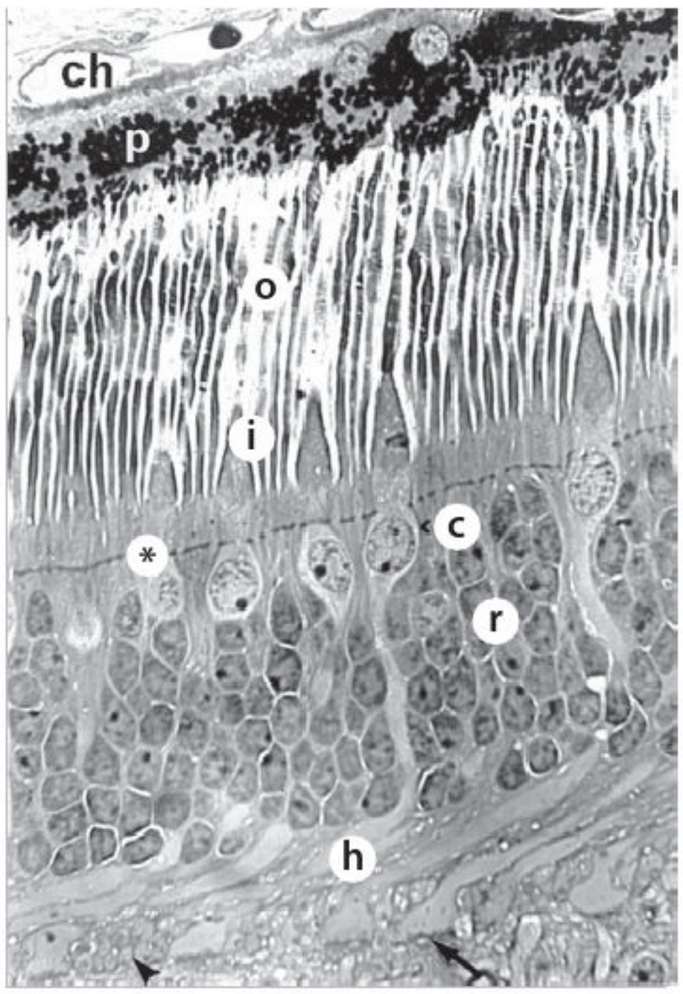

Figure 2.6 Human retina showing the larger, oval cell bodies of the cones (c) with lighter nuclei lying adjacent to the outer limiting membrane (*). They are clearly distinguishable from the smaller, rod cell bodies (r) with more densely staining nuclei, which are more densely packed and form several layers at this retinal location. Cone and rod inner fibres (h) terminate respectively in pedicles (*arrow*) and spherules (*arrowhead*). ch, choriocapillaris; p, retinal pigment epithelium; i, inner segment; o, outer segment.

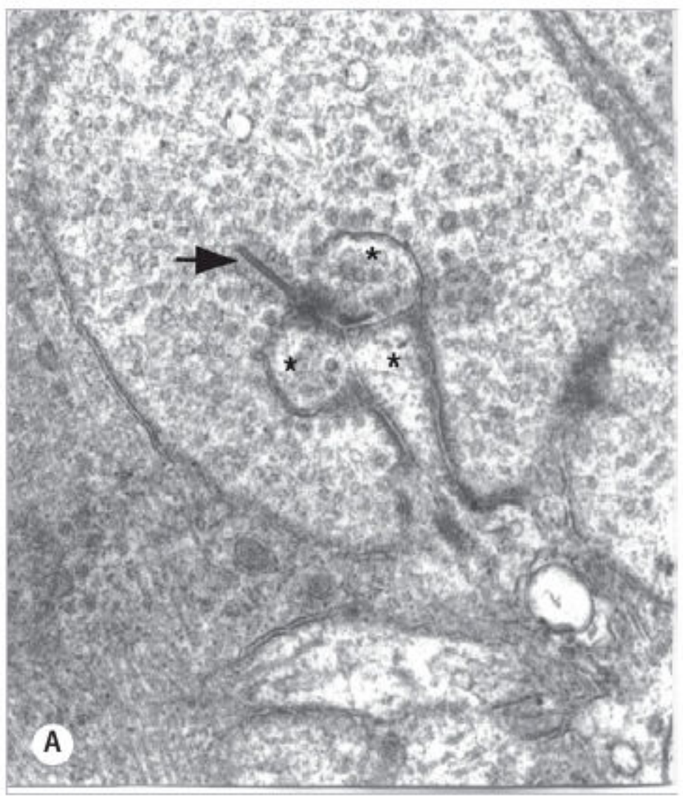

Figure 2.7A Electron micrograph of a transverse section through a rod spherule showing three distinct invaginating profiles (*) and a synaptic ribbon (*arrow*).

Not all synaptic contacts are confined within invaginations of the terminals. Cone pedicles and bipolar cell dendrites also make flat contacts (Fig. 2.7C), although there appear to be no flat contacts on spherules. These superficial synapses resemble conventional synapses with presynaptic intramembranous particles and postsynaptic densities, but are virtually devoid of closely adjacent vesicles, suggesting a synaptic mechanism distinct from that at the invaginating ribbon synapses. Nevertheless, both types of synaptic specialization represent an initial

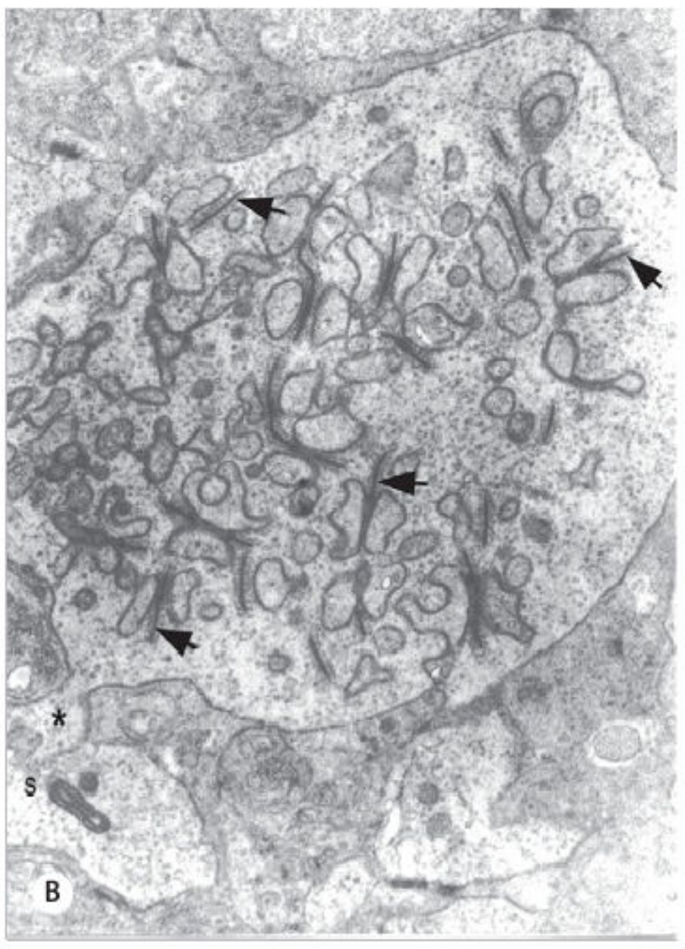

Figure 2.7B Electron micrograph of a cone pedicle sectioned parallel and close to its base, showing approximately 25 synaptic ribbons (*some indicated by arrows*) each with aligned vesicles. In the bottom left-hand corner, a pedicle process (*) appears to be contacting a rod spherule (s).

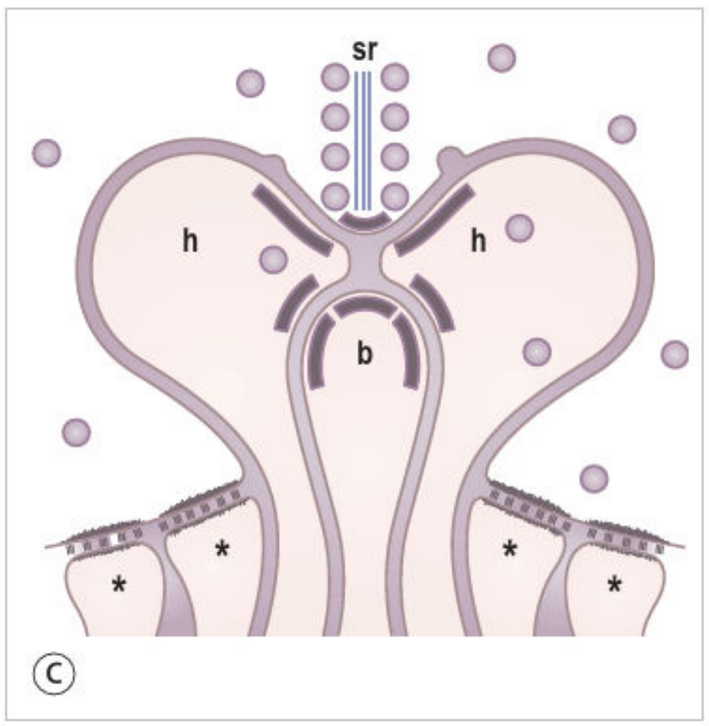

Figure 2.7C Schematic of the synaptic arrangements at a cone pedicle. Two horizontal cell dendrites (h) and one dendrite from a bipolar cell (b) invaginate the base of the pedicle, forming a triad. The ridge of the invaginations is directly opposite the synaptic ribbon (sr) of the pedicle. Synaptic vesicles align either side of the ribbon and the density is located at the base of the ribbon adjacent to the presynaptic membrane. In addition to the invaginating processes, the location of flat bipolar contacts with the pedicle are indicated (*).

locus in the visual pathway for an influence of lateral information transmission on the vertical path of information transmission through the retina.

Postreceptor organization

Vertical interactions

Information passes from the photoreceptors to the second-order neurons, the bipolar cells, at the cone pedicles and rod spherules in the outer plexiform layer. Subsequently,

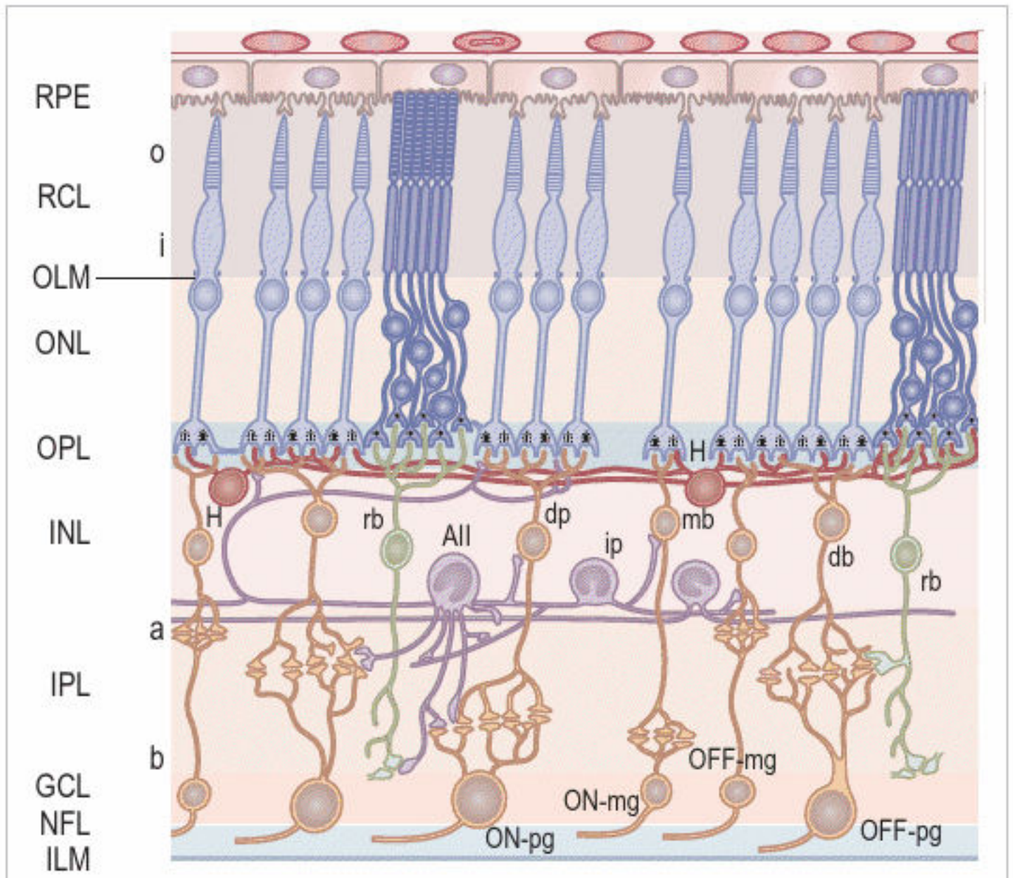

Figure 2.8 A schematic showing the morphological variety of retinal neurons and their connectivities in the context of the histologically defined layers. A number of specific cell types are highlighted: see text for details. Laminar conventions: RPE, retinal pigment epithelium; RCL, rod and cone layer (o, outer segment; i, inner segment); OLM, outer limiting membrane; ONL, outer nuclear layer; OPL, outer plexiform layer; INL, inner nuclear layer; IPL, inner plexiform layer (a, sublamina a/OFF layer; b, sublamina b/ON layer); GCL, ganglion cell layer; NFL, nerve fibre layer; ILM, inner limiting membrane. Cell types: AII, AII amacrine cell; H, horizontal cell; mb, midget bipolar; db, diffuse bipolar; rb, rod bipolar; ip, interplexiform cell; ON-pg, ON-type parasol ganglion cell; OFF-pg, OFF-type parasol ganglion cell; ON-mg, ON-type midget ganglion cell; OFF-mg, OFF-type midget ganglion cell.

with one significant exception that will be addressed further below, information passes from bipolar cells directly to the third-order neurons, the ganglion cells, at the inner plexiform layer (**Fig. 2.8**). This vertical route will be discussed before directing attention to the horizontal and amacrine cell populations that comprise the lateral pathways for information flow in the retina.

Bipolar cells

In the pathway from photoreceptors to bipolar cell to ganglion cell, glutamate is the neurotransmitter at the respective synapses. Nevertheless, the distribution of different post-synaptic glutamate receptors determines that at the photoreceptor–bipolar cell synapse the visual signal is channelled into distinct ON and OFF pathways, this terminology reflecting the response of a cell's receptive field centre to either increasing or decreasing increments in luminance (Werblin and Dowling 1969).

Throughout the retina the connections within the ON and OFF pathways remain largely segregated. At the pedicles, cone ON bipolar cells make contacts within the invaginating synapses of the pedicles, while cone OFF bipolar cells make contacts mainly at the superficial synapses of pedicles (see **Fig. 2.7C**) (Wässle 2004).

Of the estimated 25 million bipolar cells in the human retina (Oyster 1999), Kolb et al (1992) defined nine distinct types of bipolar cell, one type specifically a rod bipolar cell and the remaining eight associated with cones. Their classification of cone bipolar cells, based on morphology and connectivity, describes various diffuse bipolar cells, flat and invaginating midget bipolar cells, and cone-specific bipolar cell types. It is now generally considered that all mammalian species have a dozen bipolar cell types, all probably having ON and OFF subtypes (Nelson and Kolb 2003).

Most of the cone bipolars are diffuse and may contact between 5 and 20 cones. The so-called midget bipolar cell pathway (named because of the ultimate connections to midget ganglion cells) is, however, characterized by a much lower ratio, especially in the central retina, where cone bipolar cells outnumber cone photoreceptors and where there is at least a one-to-one relationship (**Fig. 2.8**). Given the different wavelength specificities of distinct cone types, the cone pathway in the central retina is therefore adapted for high-resolution spatial detail and colour. In the perifoveal and further peripheral retina a greater number of cones contact each midget bipolar cell, except in the case of the blue cone bipolar, which contacts only one or two blue cones (Kouyama & Marshak 1992).

The solitary rod bipolar cell type is an ON bipolar cell and the number of rods connected to a bipolar cell is greater than the cone bipolar cell ratio (**Fig. 2.8**). Each rod bipolar cell contacts from 20 to 80 rod spherules, lower numbers being characteristic of the rod bipolar cells in the central retina, with higher numbers peripherally. The rod pathway is therefore a convergent pathway. The implication of this arrangement is that, unlike the cone pathways in the central retina, the rod pathway pools inputs from many receptors, diminishing the amount of information relating to detailed spatial form but enhancing the probability of light detection, and consequently underlying the rod system's role in scotopic conditions.

Bipolar cell processes maintain the segregation of ON and OFF subtypes, making their synapses within specific sublaminae of the inner plexiform layer. OFF and ON cone bipolar synapses are segregated, respectively, to sublaminae a and b (**Fig. 2.8**). The bipolar terminal, like the photoreceptor terminal, possesses a specialized ribbon synapse. In this case, commonly there are two postsynaptic elements comprising a dyad, one being a ganglion cell dendrite, the other an amacrine cell process.

Ganglion cells

The retinal output is carried ultimately by the ganglion cells. Unlike most other retinal neurons, ganglion cells generate action potentials, other cell types generally operating via local, graded potentials. Various classifications have been reported, some based on morphological features such as cell body size and characteristics of their dendritic arborization, while other schemes have emphasized the physiological properties of different ganglion cell types. The focus here is on the former, which represent a less ambiguous, definitive description. Further, studies of the macaque retina will also be central (e.g. Leventhal et al 1981; Perry et al 1984; Rodieck 1988; Rodieck & Watanabe 1993).

The median number of ganglion cells in the human retina has been given as 1.12 million (Curcio & Allen 1990) and 18 different types have been described within this population (Kolb et al 1992). The cell bodies of these neurons are located in the ganglion cell layer of the retina, varying in density from the macular region, where, outside the

foveola, they can be up to about 10 cells thick, decreasing to one cell thick at the peripheral margin.

Particular morphological types with different patterns of dendritic branching within the inner plexiform layer were initially described by Cajal (in Polyak 1941). Given the likelihood that the distinct branching patterns indicate inputs from different bipolar and/or amacrine cell populations, the clear implication is that the distinct cell types also exhibit different physiological, and consequently functional, properties.

On the basis of their dendritic morphology, Polyak (1941) distinguished parasol and midget ganglion cell types. The midget cell population represents by far the largest ganglion cell type, at about 80% of the total, while parasol ganglion cells make up 10% of the total, both types being distributed across the retina (Perry et al 1984). The midget cell type, as the name implies, possesses a small cell body and fairly compact dendritic tree, but can be further classified as two subtypes on the basis of distinct levels of dendritic stratification in the outer and inner sublaminae of the inner plexiform layers. The parasol cell population has a larger cell body and a larger dendritic expanse than the midget cells at any location on the retina and can also be subtyped on the basis of dendritic stratification: in this case to two different sublaminae within the central zone of the inner plexiform layer (**Fig. 2.8**) (Watanabe & Rodieck 1989). These different levels of dendritic stratification reflect the continued segregation of ON and OFF pathways through the retina, ON cone bipolars making synapses with ON midget or parasol ganglion cells, and OFF cone bipolars with OFF ganglion cells (Kolb & Dekorver 1991; Dacey & Lee 1994). Cone bipolars are therefore intermediates in a direct path from cone photoreceptors to the ganglion cell output from the retina.

However, the rod pathway to ganglion cells is organized differently; rod bipolar cells receive their input from rod photoreceptors and their processes arborize in the inner, ON sublamina of the inner plexiform layer. Here, rather than synapse directly with ganglion cells, the rod bipolar cells connect to the processes of AII type of amacrine cell (**Fig. 2.8**), which pool the inputs from a number of rod bipolars. The rod pathway then joins the cone pathway via gap junctions with the processes of ON cone bipolar cells and GABAergic synapses with OFF cone bipolar cells. These then connect respectively to ON and OFF ganglion cells (DeVries & Baylor 1995).

It seems likely that this is not the only route via which the rod signal influences the cone pathways. The human electroretinogram suggests there may be two rod pathways, a slow path and a faster path. The slow path may reflect the rod bipolar to AII amacrine circuitry, whereas the fast path could be the direct entry of the rod signal into the cone path at the outer retina (Stockman et al 1995). The lateral gap junction connections found between rod spherules and cone pedicles could represent the route for rod responses in cone photoreceptors (Schneeweis & Schnapf 1995).

Midget ganglion cells in the central retina receive their input from bipolar cells which themselves receive input from a single cone (Kolb et al 1992; Dacey 1993a; Calkins et al 1994; Kolb & Marshak 2003). This midget system seems to represent a 'private-line' arrangement in its path to the brain, conveying information relating to specific wavelengths with high acuity. In contrast, parasol cells receive inputs from a number of diffuse bipolars, which receive their input from several photoreceptors (Jacoby et al 2000). These different circuitries imply that the midget and parasol systems differ in their functional properties. This is supported by the observations that the parasol input is divided, with approximately 20% from bipolar cells and 80% from various amacrine cells (Martin & Grünert 2003), while midget cell input appears to be split 50:50 between bipolar and amacrine cell input (Kolb et al 1992). As will be discussed below, these two types of ganglion cell, which together represent 90% of ganglion cells, send their axons to the primary thalamic relay, thus conveying their signal to the primary visual cortex, and represent the neural substrate of the majority of the 'dimensions' of vision.

The remaining 10% of the ganglion cell population comprises a relatively large number of different types, distinguished mainly by their dendritic morphology (e.g. Kolb et al 1992; Peterson & Dacey 2000; Telkes et al 2000). Within this population, the small-field bistratified ganglion cells have received particular attention because these cells, like the midget and parasol types, send their axons to the main retinorecipient thalamic relay nucleus. This cell type, with a dendritic expanse slightly greater than parasol cells, has been described in both monkey and human retina (Dacey 1993b) and appears to correspond to the blue-ON ganglion cell type (Dacey & Lee 1994).

One final type of ganglion cell must be given particular mention. This type, in which the dendritic tree gives the cell its description as a widefield diffuse ganglion cell, is of interest because it is intrinsically photosensitive, and thus represents an inner retinal photoreceptor. There are only about 3000 of these cells in the primate retina (<0.5% of total ganglion cells), with a dendritic tree spread among the largest found, and they express a melanopsin photopigment (Dacey et al 2005). In rodents, these cells have been considered the irradiance-dependent retinal substrate for the pupillary light reflex and for entraining circadian rhythms (Hattar et al 2002). Recent evidence also suggests an involvement of these cells in the primate pupillary light reflex (Gamlin et al 2007), but it is questionable whether the central projections from these cells can be considered a parallel, 'non-image forming' pathway, distinct from those originating at rods and cones (Dacey et al 2005).

Lateral interactions

Horizontal cells

There are at least two types of horizontal cell in the primate retina, H1 and H2, which represent lateral influences on retinal transmission in the outer retina (**Fig. 2.8**). Both types have a maximum density at the foveal margin, but the H2 population is outnumbered by twice the number of H1 cells in the peripheral retina and fourfold at their respective peaks (Wässle et al 2000). Their connections at the cone and rod ribbon synapses provide feedback to rods and cones as well as bipolar cells, with the likely outcome that the presence of a local signal is enhanced relative to more distant inputs. The H1 group has been considered by some to be heterogeneous, including an H3 subdivision (Kolb et al 1992).

The H1 cell type has a long axon terminating in a clustered arborization while the H2 type has a sparse spread of arbors at the ends of its axon (Boycott & Kolb 1973). Within each

population the cells are coupled to each other via gap junctions (Dacey et al 1996). However, the two types differ in their photoreceptor connections. The dendritic field of the H1 type contacts red and green cones, whereas the axon terminals connect to rods as the lateral components of the triad in rod terminals. In contrast, the H2 type of horizontal cell connects in particular to blue cone pedicles (Boycott & Kolb, 1973; Ahnelt & Kolb 1994a, b; Dacey et al 1996). The axon, rather than providing a basis for integrating the signals from the dendritic and axonal extremes, has the effect of electronically isolating the dendritic and axonal fields, so these two subsets of horizontal cells can be viewed as primarily processing either blue or red and green signals.

Amacrine cells

Amacrine cells are the substrate for lateral influences in the inner retina (**Fig. 2.8**). A large proportion of synapses on ganglion cells are from amacrine cells (Calkins et al 1994; Jacoby et al 1996), which also input onto the terminal processes of bipolar cells.

Most primate amacrines use either GABA or glycine as a neurotransmitter and may be categorized accordingly (Kalloniatis et al 1996). The majority appear to be glycinergic (Crooks & Kolb 1992; Koontz et al 1993) but the proportion of GABAergic amacrine synapses (80–90%) is much greater (Koontz & Hendrickson 1990), possibly reflecting a greater density of dendritic ramification by GABAergic amacrine cells. Nevertheless, regardless of their relative synaptic distributions, the inhibitory properties of both GABA and glycine appear to result in an amacrine cell circuitry that generally provides negative feedback on to bipolar cell terminals and inhibitory feedforward on to ganglion cells.

There are approximately 25 types of amacrine cell in the primate retina and they are the most morphologically variable retinal cell group (Mariani 1990; Kolb et al 1992). On the basis of their dendritic spread they can be classified into narrow/small-, medium-, or wide/large-field amacrine cells. Most amacrine cell bodies are located in the inner nuclear layer (**Fig. 2.8**), but they may also be found in the inner plexiform layer and in the ganglion cell layer. In the latter case they are often referred to as displaced amacrine cells, and in the peripheral retina they may even outnumber the ganglion cell population in that layer (Curcio & Allen 1990).

Although the vast majority of bipolar and ganglion cells stratify in either the ON or OFF sublaminae of the inner plexiform layer, roughly half of the amacrine cell population appear to have dendrites that arborize in both the ON and OFF sublaminae (Kolb et al 1992). Such bilaminar stratification is characteristic of the AII amacrine cell type, which was discussed above in relation to the vertical retinal pathway for the rod signal (**Fig. 2.8**). This small-field, glycinergic amacrine cell type also contains the calcium-binding protein calretinin, and their peak density is approximately 5000 cells/mm^2 in the perifoveal retina (Wässle et al 1995).

Many amacrine cells co-localize GABA or glycine with another neurotransmitter. A well-characterized example is the starburst amacrine cell, a medium-field type which also contains acetylcholine (Rodieck 1989; Rodieck & Marshak 1992). They also comprise ON and OFF subtypes. The cell bodies of the OFF-type cells are found in the innermost division of the inner nuclear layer and, as might be expected, their dendrites ramify in the outer, sublamina a, of the inner plexiform layer. The ON-type are included in the displaced amacrine cell group, their cell bodies located in the ganglion cell layer and their dendritic tree arborizes in the inner, sublamina b, of the inner plexiform layer (Rodieck & Marshak 1992).

In Cajal's original descriptions, amacrine cells were named as such because of their apparent lack of an axon (amacrine = axon-less). However, more recently, multiple axonal processes have been identified in a variety of large-field amacrine cells. One of these is the GABAergic, AI amacrine cell, which can, in the central retina, have a dense dendritic tree of about 0.5 mm in diameter but a polyaxonal arborization that can extend to, and maybe even beyond, 4.5 mm. In addition, unlike retinal neurons in the outer retina, these amacrine cells generate action potentials (Stafford & Dacey 1997).

Axon-like processes are also found in a dopamine-containing GABAergic amacrine cell type, some extending up to 5 mm from the cell body. The extensive spread of these processes means that, although there are less than 10 000 of these cells in the retina, they nevertheless cover its entire extent (Mariani et al 1984; Dacey 1990). Some have considered that this dopaminergic amacrine cell type may also represent one form of the interplexiform cell, a cell type that has been studied most extensively in non-mammalian species. These cells have cell bodies in the inner nuclear layer along with other amacrine cells and receive synaptic inputs in the inner plexiform layer but make synaptic output connections not only in that layer but also onto bipolar and horizontal cell processes in the outer plexiform layer (**Fig. 2.8**). This represents a potential path for retinal information feedback from the inner to the outer retina. The dopamine content of these cells in some primates (Dowling 1986) may serve not only to adjust the overall sensitivity of retinal mechanisms but also to regulate the gap junction coupling of horizontal cells (Hampson et al 1994) and thereby the spatial organization of receptive fields in the neurons of the outer retina.

Retinal outflow and the optic nerve, chiasm and tract

Retinal ganglion cell axons represent the output path from the retina and they form the nerve fibre layer, bounded at its innermost extent by the inner limiting membrane, a specialization of the end feet of the Müller glial cells.

The ganglion cell axons display a characteristic path across the inner retinal surface to their exit point posteriorly at the optic disc, the visuotopic position of the blind spot due to the absence of photoreceptors at this location. Axons from nasal locations in the retina take a path directly to the disc, as do axons from superior and inferior temporal quadrants in temporal retina. Those arising from ganglion cells nasal to the fovea also course directly to the optic disc, forming the papillomacular bundle. In contrast, those arising from points temporal to the fovea take an arcuate course above or below the fovea, depending on whether they originate from inferior or superior retinal locations respectively (Hogan et al 1971).

Optic nerve head

Approximately one million ganglion cell axons exit the retina to form the optic nerve (Potts et al 1972a; Balazsi et al 1984; Sanchez et al 1986; Mikelberg et al 1989; Jonas et al 1990, 1992). In doing so, their course takes them initially through the intraocular (or intrascleral) optic nerve, a 1 mm length also referred to as the optic nerve head, which represents a region of considerable specialization. The scleral organization is modified here to form a sieve-like meshwork, the cribriform plates, which support the axons in this part of their course, designated the lamina cribrosa (**Fig. 2.9A**). The cribriform plates are composed of various extracellular matrix components. The cores of these trabeculae comprise mainly elastin with collagen types I and III, while collagen type IV and laminin are found associated with their margins (Hernandez et al 1987; Rehnberg et al 1987; Morrison et al 1989; Hernandez 1992). Various proteoglycans have also been localized to the region of the lamina cribrosa (Caparas et al 1991).

Prior to their course through the lamina cribrosa, the unmyelinated axons are gathered into astrocyte-lined fascicles. These are continuous with the pores that penetrate the cribriform plates (**Fig. 2.9B**) (Anderson 1969; Hogan et al 1971; Ruskell 1988). These pores are variable in size, and the thickness of the connective tissue plates separating the axon bundles varies accordingly. The pore sizes are apparently generally larger in superior and inferior quadrants at the lamina cribrosa (Quigley et al 1990), suggesting that the physical support of axons is weaker in these locations, and hinting that there may be an increased likelihood of axons in these locations being disrupted by elevated intraocular pressure.

In the postlaminar region of the optic nerve head, the ganglion cell axons are myelinated, accounting for the increase in the nerve diameter from 1.5 mm at the laminar level to approximately 3–3.5 mm. Given the central neural origin of the optic nerve, the myelin sheath of the axons derives from oligodendrocytes, while the general absence of myelination along the prior course of the axons has been ascribed to a possible barrier to the migration of oligodendrocyte precursor cells by the lamina cribrosa (Perry & Lund 1990).

It should be noted, prior to considering further proximal locations, that not all axons in the optic nerve originate from retinal ganglion cells. A few, maybe only a dozen, are centrifugal axons originating from cell bodies in the posterior hypothalamus and dorsal raphe nucleus. They arborize in the inner retinal layers and possibly function to regulate retinal interneuron activity and blood flow (Gastinger et al 2006; Repérant et al 2006).

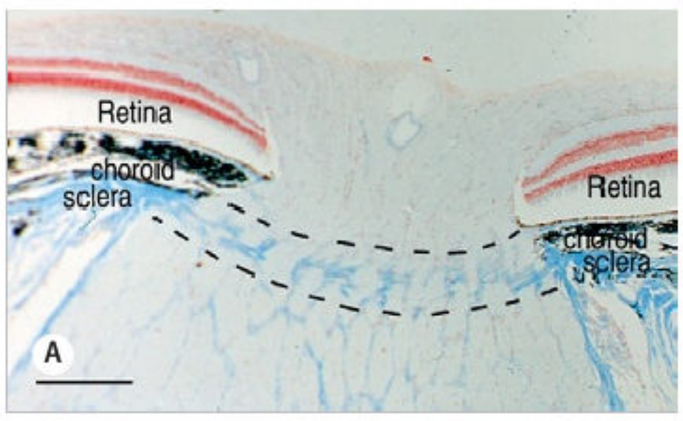

Figure 2.9A Section through the optic nerve head. The location of the lamina cribrosa is highlighted by the blue-stained plates extending through the nerve head at the level of the sclera (*indicated by broken lines*). Scale bar 0.5 mm.

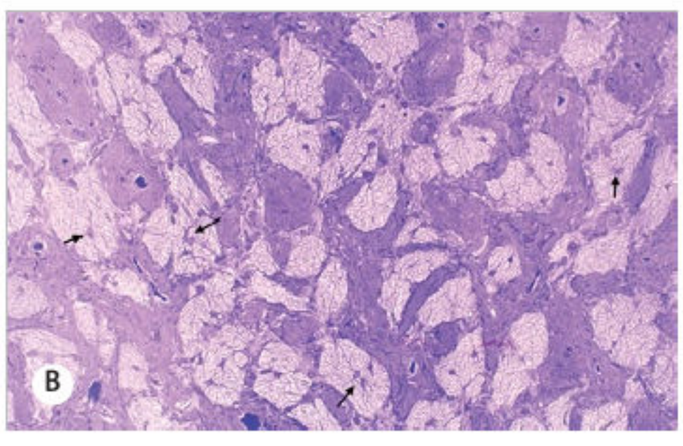

Fig. 2.9B Transverse section of the optic nerve at the peripheral margin of the lamina cribrosa. The lighter stained regions represent the pores occupied by ganglion cell axons, the darker regions being interfascicular tissue. The arrows indicate the cell bodies of astrocytes amongst the axon bundles.

Postlaminar optic nerve

The optic nerve at the postlaminar level is invested by a meningeal sheath; the dura mater, an outer, thick collagenous layer of connective tissue, envelops the arachnoid mater, a middle layer of trabeculae with collagen cores surrounded by meningothelial cells. The innermost layer, the pia mater, is a delicate connective tissue layer in which are embedded numerous blood vessels. Connective tissue septa of pial origin extend into the nerve to surround the axon fascicles at this postlaminar level of the nerve (**Fig. 2.9C**). The spaces between the meningeal layers, the subdural and subarachnoid spaces, are filled with cerebrospinal fluid. The latter is continuous with the intracranial subarachnoid space, meaning that increased intracranial pressure has the potential for direct compression on the optic nerve.

The optic nerve carries the myelinated retinal ganglion cell axons from the intraocular segment to the optic chiasm, a distance of 40–50 mm, which can be subdivided into three further segments. Firstly, the intraorbital part of the optic nerve is about 20–30 mm in length and extends to the optic foramen (canal). The path is not direct but arcs laterally near the orbital apex, resulting in about 6 mm of slack that enables compliant movement of the nerve during ocular rotation (Wolff 1948). Where the nerve enters the foramen the dural sheath fuses with the orbital periosteum, limiting potential movement along the subsequent intracanalicular part of the nerve. The path of the nerve through

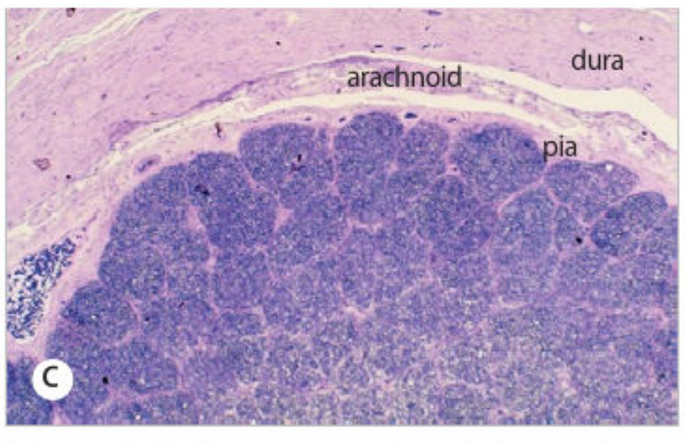

Figure 2.9C Transverse section of the postlaminar optic nerve showing the fascicular organization of the nerve and the location of the different meningeal layers.