Published in final edited form as:

Trends Neurosci. 2014 October ; 37(10): 594–603. doi:10.1016/j.tins.2014.07.009.

Neuronal remodeling in retinal circuit assembly, disassembly and reassembly

Florence D. D'Orazi*, Sachihiro C. Suzuki*, and Rachel O. Wong[#]

Dept. of Biological Structure, University of Washington, 1959 NE Pacific St., Seattle, WA 98195

Abstract

Developing neuronal circuits often undergo a period of refinement to eliminate aberrant synaptic connections. Inappropriate connections can also form amongst surviving neurons during neuronal degeneration. The laminar organization of the vertebrate retina enables synaptic reorganization to be readily identified. Synaptic rearrangements are shown to help sculpt developing retinal circuits, although the mechanisms involved remain debated. Structural changes in retinal diseases can also lead to functional rewiring. This poses a major challenge to retinal repair because it may be necessary to untangle the miswired connections before reconnecting with proper synaptic partners. Here, we review our current understanding of the mechanisms that underlie circuit remodeling during retinal development, and discuss how alterations in connectivity during damage could impede circuit repair.

Keywords

retina; synapses; circuit refinement; retinal degeneration

Making and breaking connections

The assembly of neuronal circuits during development involves cellular and molecular processes that are orchestrated at multiple levels. Component cell types need to be generated in the right numbers and positioned at their final locations. Each cell type must then elaborate axons and dendrites to connect with suitable synaptic partners, and establish the mature number, type and distribution of synapses. Also, synaptic machinery has to be localized appropriately, and the molecular composition unique to each synapse type must be attained to ensure proper function. Often, these developmental events involve several steps of refinement before circuits are fully established. For example, axonal and dendritic arbors may undergo pruning to achieve their final patterns, and the molecular compositions at synapses may be reconfigured before maturation. Here, we review our current understanding of the cellular strategies and molecular mechanisms that underlie the structural and

© 2014 Elsevier Ltd. All rights reserved.

[#]Corresponding author: Wong, R.O. (wongr2@uw.edu).

*These authors contributed equally

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final citable form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

functional remodeling events that organize circuits during development. Further, we highlight the structural and synaptic reorganization that take place in disease conditions, and consider how these rearrangements could potentially impact circuit repair. We will focus on the vertebrate retina for which organization and function are relatively well understood, and for which circuit deconstruction and dysfunction can be probed in detail using anatomical and physiological techniques.

Remodeling and retinal circuit assembly

Organizing synaptic laminae

The laminar organization of the vertebrate retina greatly facilitates investigation of circuit development. The vertebrate retina comprises five major classes of neurons that are arranged in three cellular layers (Figure 1A,B). Synaptic connections are organized into two layers, the outer (OPL) and the inner (IPL) plexiform layers. The IPL is further subdivided into two major sublayers. Within the inner (ON) sublamina lie the processes and connections of retinal ganglion cells (RGCs), bipolar cells and amacrine cells that depolarize in response to increased illumination. Conversely, neurons that are hyperpolarized by light increments stratify their processes in the outer (OFF) sublamina. Specialized presynaptic structures called “ribbons” mediate excitatory glutamatergic transmission in both plexiform layers [1] (See Glossary) (Figure 1C,D). Disruption to the layered organization of the retina greatly perturbs its function.

Several elegant studies have identified many guidance cues and adhesion molecules responsible for confining synaptic connectivity to the plexiform layers (summarized in Box 1). Structural and functional organization within each layer, however, can undergo further refinement during development before the adult circuitry emerges. In particular, early work in cat and ferret underscored the importance of structural remodeling in constraining the dendritic stratification of ON- or OFF-RGCs to their correct sublamina [2, 3]. Initially, the dendrites of ON and OFF alpha and beta RGCs span the depth of the IPL. But, comparisons of fixed tissue across ages suggested that these RGC types constrain their dendrites within the ON or OFF sublamina after eliminating branches at inappropriate depths; failure to undergo such rearrangements leads to abnormal connectivity and function [4].

Box 1

Molecular cues regulating synaptic lamination

Repulsive signaling from the semaphorin/plexin family of transmembrane guidance cues prevents inner retinal neurons from mistargeting to the OPL by constraining their neurites to the IPL. In mouse retina, loss of semaphorins Sema5A and 5B, or their receptors PlexinA1 and A3, allows several RGC and amacrine cell subtypes to aberrantly extend their dendrites into the outer retina [17] (Figure I, *Sema5A/5B* dKO). Other members of the semaphorin/plexin family more narrowly confine the dendritic projections of RGCs to the ON and OFF sublaminae within the IPL [16], as well as mediate the segregation of ON- and OFF-starburst amacrine cell dendrites into two plexuses [18]. Notably, in *Sema6A* or *PlexinA4* mutants, melanopsin-expressing M1 RGCs form an additional arbor in the ON sublamina, matching changes in the arborization patterns of their

presynaptic partners, the tyrosine hydroxylase-expressing amacrine cells (Figure I, *Sema6A* KO). Immunoglobulin superfamily (IgSF) adhesion molecules also restrict connectivity choice, whereby synaptic partners are directed to a specific depth of the IPL as well as molecularly matched to each other within a plexus [12]. In chick, non-overlapping subsets of RGCs and amacrine cells express Dscam, Dscam-like, Sidekick-1 or Sidekick-2, and the pre- and postsynaptic partners expressing a given IgSF molecule arborize in the same IPL sublaminae via homophilic adhesion [15]. Contactins, homologs of Sidekick and Dscam, regulate the sublamina specificity of subsets of inner neurons in chick retina [14]. Although adhesion molecules constrain neurite lamination and therefore the connectivity choices of inner retinal neurons, synaptogenesis can still proceed even in the absence of these molecules. In mice lacking *Fat3*, an atypical cadherin, the dendrites of amacrine cells form ectopic synaptic layers in the INL and GCL, where, in the case of AII amacrine cells, synaptic contacts are formed with the appropriate partners albeit in the wrong location (Figure I, *Fat3* KO) [96].

In contrast, recent studies in zebrafish and mice brought into question whether large-scale dendritic pruning is adopted universally by RGCs as a primary means to attain proper lamination. Fluorescent labeling of specific RGC types in transgenic mice [5] and time-lapse imaging in zebrafish [6] together revealed that RGCs employ several lamination strategies, which rely on dendritic remodeling to varying degrees: (1) Large scale refinement of an initially diffuse arbor; (2) Fine scale refinement of an arbor that is biased to the ON or OFF layer (i.e. pruning within a layer); and sequential loss (3) or addition (4) of an entire arbor or arbors (Figure 2). It is not known why RGCs adopt diverse strategies. But, it is clear, that the extent of remodeling is not correlated with specific features of the RGCs, such as ON vs. OFF function or the number of arbors, nor is it species-dependent. Retinal neurons other than RGCs also undergo neurite remodeling to varying degrees to achieve their final lamination patterns. Mouse rod bipolar cells (RBCs) and ON-cone bipolar cells (ON-CBCs) extend axonal processes from a neuroepithelial-like stalk that eventually retracts as the axonal terminal elaborates. Lateral axonal growth is confined to the ON sublamina, suggesting that these bipolar cells do not undergo significant remodeling to target their major synaptic layer [7]. Whether fine-scale refinement occurs to further restrict the axonal terminals of the 10 or more types of mouse bipolar cells [8, 9] to their respective layer within the IPL remains unknown. Similarly, migrating amacrine cells that are initially multipolar preferentially direct neurite outgrowth towards the IPL and rapidly stratify once their somata reach their final location [10]. In contrast, rod and cone photoreceptor axons in the ferret retina transiently extend all the way into the IPL [11]. Simultaneous imaging of pre- and postsynaptic processes is now needed to ascertain how axonal and dendritic growth and remodeling are coordinated during development to establish the relevant connections.

Refining synaptic connectivity

It is clear that lamination initially limits the plethora of potential synaptic partners for any given retinal circuit [12] (Figure 2). However, even within a lamina, only some cells form stable connections with each other. Several families of adhesion and guidance molecules that serve to match synaptic partners are now known [13–18]. Nonetheless, some retinal

neurons are found to wire erroneously, necessitating synapse elimination to settle on their final partners. For example, A-type ON-RGCs are contacted by RBCs before eye-opening, but lose these contacts thereafter [19] (Figure 2). Likewise, zebrafish H3 horizontal cells initially make aberrant cone contacts, but eventually eliminate these connections in favor of UV and blue cones [20].

Synapse elimination also contributes to finalizing patterns of connectivity with presynaptic cells of the same type. Mouse ON-CBCs initially make exuberant connections with cone photoreceptors that are pruned with maturity. Time-lapse imaging in mouse retina revealed that over a protracted period of development, Type 7 and 8 ON-CBCs eliminate some of their cone connections, but Type 6 ON-CBCs undergo little to no remodeling [21]; the extent of bipolar cell dendritic refinement appears to be correlated with dendritic field size. Starburst amacrine cells initially provide balanced inhibition in both the 'preferred' and 'null' direction of direction-selective RGCs, but upon maturity, inhibition becomes greater in the null direction [22, 23]. Whether or not there is structural change underlying this functional reorganization is not yet known, it is likely to include a selective loss of inhibitory synapses from the starburst cells serving the preferred direction. These observations lead us to conclude that (i) synaptic drive is refined even between 'matched' partner types, and (ii) postsynaptic cells sharing the same population of presynaptic partners differentially employ mechanisms of remodeling to achieve their final connectivity.

Neurotransmission and circuit refinement

Synaptic refinement and neurite remodeling in developing visual CNS circuits have classically been found to rely on activity-dependent mechanisms [24, 25]. In the retina, chronic intraocular application of 2-amino-4-phosphonobutyrate (APB), an agonist of metabotropic glutamate receptors (mGluR6) (see Glossary) on ON-bipolar cell dendrites, prevented the emergence of stratified arbors of alpha and beta RGCs [3]. In line with this finding, sensory deprivation of mice resulted in an abnormal proportion of RGCs with ON and OFF responses, implying the many RGCs failed to undergo dendritic refinement [26, 27]. It is proposed that transmission acts together with CD3 ζ , a component of the Major Histocompatibility Complex (MHC) Class I receptor involved in the immune response, to shape RGC dendritic lamination [28]. Indeed, MHC Class I proteins have also been found to regulate activity-dependent synaptic remodeling in other parts of the developing visual system [29–31].

However, several studies have challenged the idea that activity-dependent mechanisms are crucial to laminar refinement. In mGluR6 knockout mice, ON-bipolar cells are hyperpolarized due to a failure of TRPM1 channels to localize to their dendritic tips [32, 33]. Despite defective transmission from these bipolar cells, ON RGC dendritic arbors stratify normally [34]. Furthermore, RGCs develop normal ON-stratified arbors when the light chain of tetanus toxin (TeNT), a protein that disrupts synaptic vesicle fusion, is expressed transgenically in ON-bipolar cells [35, 36]. The disparate findings for a role of transmission in RGC dendrite lamination may be due to species differences, distinct actions of each manipulation [37], or it may reflect separate requirements for transmission across

cell types. To distinguish amongst these possibilities, comparison of the effects of different manipulations on a given RGC type is necessary [37].

Regardless of its controversial role in dendritic lamination, it is clear that neurotransmission is not always required for synapse refinement in the retina. Suppression of transmission from ON-bipolar cells does not preclude elimination of RBC synapses with the A-type ON-RGC [19], nor does GABA_A receptor blockade disrupt the emergence of direction-selectivity in RGCs [22, 23]. It is not known whether transmission from zebrafish red or green cones is necessary for elimination of their early synapses with the H3 horizontal cell, but it is evident that transmission from the preferred partners (UV and blue cones) is not needed for this refinement step [20]. Thus, unlike higher order circuits in the visual pathway [24, 25], there is no compelling evidence for activity-dependent synaptic competition in shaping retinal circuits.

Although neurotransmission is not important for synapse elimination in the retina, it does influence synapse formation. Several types of ON-CBCs make fewer cone connections during development in dark-reared mice [38], and time-lapse imaging showed that TeNT-expressing ON-CBCs exhibit a reduced rate of synaptogenesis with the A-type ON-RGCs [35]. Neurotransmission is also integral to selectively suppressing synaptogenesis with one afferent type over another in some convergent circuits. At maturity, zebrafish H3 horizontal cells contact UV cones preferentially over blue cones, with a ratio of about 5 UV contacts to 1 blue cone contact. Transmission from the UV cones limits horizontal cell synaptogenesis with the blue cones, giving rise to the horizontal cell's UV cone-biased connectivity [20]. Thus, neurotransmission is involved in shaping connectivity of many, but not all developing retinal circuits, and acts to regulate both the formation of synapses and the elimination of connections by neurite remodeling.

Assembling the synapse

An important step in the maturation of excitatory synapses in the retina is the localization of ribbons to presynaptic sites. Thus far, it is not known how ribbons are trafficked to or assembled at the synapse during development, nor is it known whether ribbons remain anchored to the presynaptic membrane once localized. Nevertheless much work has underscored the importance of investigating ribbon synapse development because of the susceptibility of this structure to perturbations during development and in disease. For example, loss of ribbon-associated proteins, such as the presynaptic cytomatrix protein bassoon, results in "floating," unanchored ribbons in the axonal terminals of both cones and rods [39, 40](Figure 3). Floating ribbons are also observed in the cones and rods of mouse [41–43] (Figure 3) and zebrafish [44–46] mutants with neurotransmission defects. However, disruption in ribbon localization may not be due to altered transmitter release *per se* because ribbons do not float but instead accumulate in abnormal numbers at individual synaptic sites of TeNT-silenced mouse bipolar cells [35]. In mutants with floating photoreceptor ribbons, neurites from the postsynaptic partners often fail to invaginate the photoreceptor terminal (Figure 3). But, the converse is not true; bipolar cells sometimes do not invaginate photoreceptor terminals even when ribbons are properly anchored, as seen in a knockout for the synapse-associated extracellular matrix protein, Pikachurin [47] (Figure 3). Thus, the

relationship between ribbon localization and neurite invagination is convoluted, and we have yet to identify how these two events are coordinated during synapse assembly. Another level of complexity is revealed by mutants in which horizontal cells fail to invaginate the photoreceptor terminal, but RBCs are unaffected, and vice-versa (Figure 3). Invaginations of horizontal cell and bipolar cell processes thus appear to be regulated independently.

On the postsynaptic side, there are unambiguous examples of remodeling during development. Whereas mGluR6 is exclusively localized to the invaginating tips of ON-bipolar cell dendrites at maturity, receptor clusters that are not associated with cones progressively disappear with maturation, a process that is influenced by sensory drive [38]. Even after glutamate receptors are localized properly at synaptic sites, their molecular composition can change with development. For example, NR2B subunits of NMDA receptors on RGC dendrites are replaced with NR2A subunits [48]. Interestingly, two functionally distinct types of direction-selective RGCs lose NR2B subunits at different rates. The mechanisms underlying subunit changes in these cells have not yet been determined, nor is it known why there is a timing difference. Whether or not transmission is responsible for modifications of these and other glutamatergic postsynaptic sites during normal development also remains to be resolved.

Remodeling in retinal disease

The most prevalent causes of blindness are diseases in which retinal neurons degenerate. Both Age-Related Macular Degeneration (AMD), a leading cause of blindness, and Retinitis Pigmentosa (RP), the most commonly inherited retinal degeneration disorder [49], result in photoreceptor degeneration. Studies of animal RP models and human RP reveal that the degeneration of rod photoreceptors, followed by cone photoreceptor death, triggers dramatic changes in the retina [50, 51]. Changes include a variety of remodeling events, from neurite loss or delamination to novel wiring, ectopic synapse formation, and modifications of glutamatergic receptor composition and localization. However, such regressive events are not exclusive to diseases with photoreceptor degeneration. Retinal remodeling also occurs in glaucoma, in which ganglion cells die [52]. In either case, when the major input or the major output neurons of the retina are lost, degeneration triggers structural and functional changes in the remaining retinal circuits.

Losing connections

Predictably, second-order retinal neurons respond to deafferentation by dismantling their disconnected neurites. In the *rd10* mouse model [53] of an autosomal recessive form of RP [49], a missense mutation in the rod-specific β -phosphodiesterase (*PDE6 β*) gene leaves rods semi-functional until they degenerate at 3 postnatal weeks, thereafter leading to cone death by 6 months of age [54–56]. Horizontal cells and RBCs retract their dendrites at the peak of rod death, and CBCs follow suit at later stages, during cone photoreceptor degeneration [54, 56–58] (Figure 4A). Clearly, the preferred afferent types are important for the maintenance of the bipolar cells' dendrites, even though both bipolar cell types elaborate their dendrites in the OPL in the absence of photoreceptor partner types during retinal development [59].

Dendrites of retinal neurons can undergo dramatic loss even when the afferent population is intact. RGC dendritic arbors shrink in primate and mouse models of glaucoma, prior to cell death [60, 61]. Alterations to synapse density and light-driven responses occur prior to noticeable dendritic retraction [60]. The early signaling events that trigger dendritic retraction in RGCs remain unknown, but it represents an interesting case in which the postsynaptic elements are dismantled without apparent retraction of the axonal terminals of the presynaptic partners [62]. Moreover, the rate of arbor shrinkage differs across RGC types [60, 61, 63], indicating that some RGC types succumb to perturbation more rapidly.

Miswiring with new partners

Neurons that have lost afferent input have also been found to remodel their connectivity. For instance, during the period when cones are still present following rod death in *rd10* mice, RBCs form novel, albeit non-invaginating, contacts with cones [54, 58] (Figure 4A). In pig and rat retinas with rod degeneration, RBCs similarly rewire with intact cone photoreceptors, and even make invaginating synapses [64, 65]. Manipulations of photoreceptor composition have previously demonstrated that bipolar cells are capable of forming connections with a novel partner type in the absence of their preferred partners during development. CBCs connect with rods in 'coneless' mice [59], whereas RBCs, as well as horizontal cells, contact cones in 'rodless' mice [66]. Such novel bipolar cell wiring patterns are also observed in mice in which either the cones or rods fail to transmit, at stages prior to photoreceptor death [67]. However, when transmission is perturbed in all photoreceptors, bipolar cells do not seek new synaptic partners [67]. Thus, although the mechanisms that direct the selection of photoreceptor partners are not yet well-defined, bipolar cell dendritic rewiring seems to depend at least in part on the presence of an active afferent.

Same partners, wrong location

A striking remodeling phenotype seen in human RP as well as in AMD is the sprouting of second-order neurons following photoreceptor degeneration [68, 69]. Although less pronounced, horizontal cell and bipolar cell neurite sprouting is also found in *rd10* mice [54] and in mice mutants lacking key presynaptic proteins (Figure 3B). Sprouting of horizontal cell neurites is more evident in the *rd1* mouse model of RP [70–72], but because rod degeneration begins during synaptogenesis, the specific cause of these remodeling events is unclear. Delamination of horizontal and bipolar cell processes does appear to be a hallmark of mouse mutants and disease models in which photoreceptors are dysfunctional but do not die. In animals with impaired photoreceptor transmission such as the *cacna1f* mutant, *nob2* [42, 73] and the CNGA3/CNGB1 double-knockout mutant [74], horizontal cell and RBC processes initially target the OPL correctly, but later sprout (Figure 4A). Intriguingly, CBCs appear less susceptible to sprouting compared to RBCs, except when horizontal cells are ablated in adult mouse retina [75, 76]. Although it is not yet understood why dendritic sprouting is more commonly observed for RBCs compared to CBCs, this finding suggests that the repair of rod and cone pathways may require distinct strategies.

What initiates the sprouting of horizontal and bipolar cell processes in retinas with photoreceptor dysfunction? Delamination appears to occur primarily when presynaptic

function is impaired, but not when postsynaptic components are altered [34, 77, 78], with the exception of the *nob4* mutant [79]. Thus, disruption of neurotransmission at OPL synapses may be necessary, but not sufficient, to trigger bipolar cell and horizontal cell neurite sprouting into the ONL. Calcium homeostasis may also play a role in neurite sprouting; perturbations of calcium homeostasis in photoreceptor terminals are correlated with retraction of rod axons into the ONL [41, 42, 80]. Observations at different ages in *nob2* mice in which photoreceptors have diminished calcium influx, suggest that rod axons retract prior to bipolar cell dendritic sprouting, and that the sprouted dendrites fasciculate with ectopic horizontal cell neurites in the ONL [42, 74]. These results support a possible role for either rod axon retraction or horizontal cell sprouting to induce bipolar cell dendritic sprouting. Most likely, rod axon retraction stimulates bipolar cell neurite growth, because changes in horizontal cell lamination do not necessarily induce dendritic sprouting in bipolar cells [81, 82]. Horizontal cell processes sprout but bipolar cell dendrites remain laminated in a mutant in which horizontal cells lack a synaptic adhesion molecule receptor, NGL-2 (Figure 3B) [82]. Also, RBCs are found to send ectopic processes into the ONL, contacting retracted rod photoreceptor axons when some horizontal cells fail to reach the OPL [76] or are ablated in adult retina [75]. Thus, the sprouting of neurites from different second-order neuron populations appears to be independently regulated during retinal disassembly.

Synapse remodeling

Reorganization of transmitter receptor localization and expression is also evident following physical and functional deafferentation of retinal circuits. Coincident with RBC dendritic retraction from the OPL after rod degeneration, mGluR6 rapidly disappears from RBC dendrites, and can become mislocalized to the soma and axons [56, 57] (Figure 4A,B). Patch-clamp recordings of individual RBCs after mGluR6 reorganization have however, produced disparate conclusions as to whether these cells retain glutamate sensitivity [57, 58]. Like the RBCs, ON-CBCs lose their mGluR6-mediated responses during cone degeneration in *rd10* mice [58] (Figure 4B). In contrast, at this stage, OFF-CBCs appear to retain their ionotropic glutamate receptor (iGluR)-mediated signaling [58] (see Glossary), despite evidence that they may remodel their dendrites [56, 57] (Figure 4B). Thus, mGluR6 maintenance on ON-bipolar cell dendrites may depend on synaptic input, whereas OFF-CBCs may retain their ability to depolarize in response to glutamate even in late stages of photoreceptor degeneration. In general, the OFF pathway seems less susceptible to disease-invoked remodeling. Further support comes from findings that ON-bipolar cells undergo functional reprogramming when mGluR6-mediated signaling is diminished. In vitro excitation mapping using reporters of glutamate receptor activation in mouse and rabbit RP models, as well as in human RP, demonstrates that ON-CBCs [83] and RBCs [84, 85] aberrantly express iGluRs after rod death and prior to cone degeneration (Figure 4B).

Additional evidence that secondary neurons may undergo functional modifications comes from cases of delamination or re-wiring. Horizontal cell and RBC neurites that extend beyond the OPL sometimes form ectopic contacts with rod terminals that have withdrawn into the ONL [42], or even onto rod cell bodies [74] (Figure 4A). These ectopic synapses frequently display many features of a functional synapse, including the presence of ribbons, as well as invaginating processes bearing glutamate receptors [42, 74]. However, ectopic

synapses can show ribbon reorganization. Electron microscopy in transmission defective mutants revealed that presynaptic ribbons present at the ectopic synapse (Figure 4A) are sometimes malformed [42], thus bringing into question how well these contacts function. Nonetheless, bipolar cell synaptic function may persist despite re-wiring. In transmission mutants in which ON- and OFF-bipolar cells re-wire with novel photoreceptor partners, the new synapses still contain the appropriate glutamate receptor types (mGluR and iGluR, respectively) [67].

Finally, cell death in the outer retina can also impact synapses in distal circuits in the inner retina, and vice versa. RGC death in mouse models of glaucoma indirectly alters the shape and number of ribbons in photoreceptor terminals [86, 87]. Likewise, photoreceptor degeneration leads to a decrease in ribbon number in CBC axons [88] and changes in RGC dendritic morphology [89]. The origins of long-range effects are not well understood, but these events nonetheless emphasize the fact that changes in connectivity in one synaptic layer do not remain confined.

Challenges to circuit repair after remodeling in disease

There are two major approaches to replace lost neurons in retinal disease: replacement via cell transplantation [90–92], or induction of cell regeneration or transdifferentiation within the adult retina [93, 94]. Stem cell approaches are increasingly successful in generating different types of retinal neurons, as are techniques to ensure survival of both stem cells [90, 91] and differentiated cells [92] after transplantation. The challenge now is that these seeded neurons must be integrated into the surviving circuitry to re-establish lost connections. A recent study demonstrated that embryonic and neonatal RGCs seeded into the adult retina integrate into the ganglion cell layer and project neurites in the IPL, where synapses may be formed [95]. How well transplanted neurons reinstate their detailed circuitry and functions remains to be elucidated. Full recovery of the normal circuitry is likely to be challenging because neurite organization, synapse structure and connectivity sometimes undergo extensive remodeling during disease (Figure 4). Moreover, some of the molecules that guide neurite growth and stratification during development are absent at maturity [17, 96]. These challenges are also inherent in approaches focused on triggering regeneration pathways within the diseased retina [93].

Finally, although replacing cells in the diseased retina is likely to become routine in the future, full recovery of function also requires rectifying changes distal to the disconnected local circuit. In addition, bystander cell death spurs the need for early intervention prior to further alterations of retinal circuits as the disease progresses [69]. Thus, studies detailing the disassembly and restructuring of circuits in retinal disease will no doubt provide valuable guidance for preventing or dismantling miswired connections in favor of establishing proper connectivity during repair.

Comparisons of retinal remodeling in development and disease

Understanding the extent of early circuit “plasticity” may provide insight into the constraints on remodeling at maturity. In the outer retina, selecting synaptic partners is not necessarily limited to early development, because re-selection of partners occurs in disease. Bipolar

cells appear capable of switching photoreceptor type partners both during development and in the adult [66, 67]. Not all retinal cell types are, however, 'exploratory' during development. Whereas some bipolar cell types undergo developmental refinement in their connectivity, some do not. Type 7 bipolar cells that do remodel during development [21], however, also demonstrate plastic changes in disease [88]. Conversely, retinal neurons that do not normally undergo extensive developmental refinement can exhibit significant structural and functional rearrangements when their major afferents are ablated [36]. Notably, in the absence of their dominant ON-bipolar cell partners, A-type RGCs that directly target their dendrites to the ON sublamina during development elaborate dendrites to the OFF sublamina, where they form novel connections with a specific-OFF bipolar cell type [36]. Thus, retinal neurons that do not exhibit much developmental remodeling appear capable of unusual plastic changes when challenged.

Because remodeling events in development and in disease share common features, knowledge of the cellular and molecular mechanisms that underlie developmental rearrangements may also help prevent restructuring in disease. For example, molecular and cellular interactions underlying dendritic retraction or pruning in developing RGCs may be involved in dendrite loss in disease. In both cases, dendrites that are eventually lost already had synaptic contacts [60], raising possibility that altered transmission triggers dendrite retraction. This seems unlikely because suppression of transmission from all bipolar inputs onto A-type RGCs does not alter their dendritic arbor size [35]. Delineating the molecular mechanisms that control dendritic retraction in development may help us to target subsets of genes that are responsible for dendritic loss in disease. Approaches akin to those that have identified key regulators of dendritic patterning and disassembly during metamorphosis in developing *Drosophila* may prove pertinent in preventing mis-patterning in disease [97].

Although much remains to be answered, we end by posing several specific questions that emerged from considering retinal circuit remodeling in development and disease (Box 2). In moving forward, it is important to remember that the extent of remodeling in the developing or damaged retina varies across circuits, and across species. Thus, it is unlikely that a single animal model system will provide all the answers. Comparative studies across species should therefore be encouraged; for example, addressing why lower vertebrates [93, 98], and not mammals, are able to regenerate their retina. By combining work across model systems and applying a multidisciplinary approach, we may more rapidly learn how to prevent regressive events during degeneration or perhaps, to make use of the remaining circuitry to reproduce essential visual functions.

Box 2

Outstanding questions

1. Why do some circuits require major refinement during development, and others do not?
2. Are molecular mechanisms and cellular strategies engaged in neurite development re-deployed during circuit reassembly?

3. What factors render some retinal neurons more susceptible to remodeling during disease?
4. How does perturbation to dendrites affect axons in development and in disease, and vice versa?
5. What are the functional consequences of the structural remodeling events that occur in disease?
6. Does miswiring in disease hamper re-wiring during circuit repair?

Acknowledgments

We thank Takeshi Yoshimatsu, Clare Gamlin, and Mrinalini Hoon for providing critical feedback on the manuscript. This work was supported by NIH grants EY10699, 17101, 14358 to R.O.W., Vision Core Grant EY01730 to M. Neitz, the Uehara Memorial Foundation Research Fellowship to S.C.S, and an NIH training grant HD07183 (F.D.D.).

Glossary

Ionotropic glutamate receptors

Ionotropic glutamate receptors (iGluRs) are glutamate-gated ion channels, and thus regulate fast excitatory transmission at ribbon synapses in the retina. The three major classes of iGluRs include N-methyl-D-aspartate (NMDA) receptors, alpha-amino-3-hydroxy-5-methyl-4-isoxazole propionic acid (AMPA) receptors, and kainate (KA) receptors. Whereas NMDA receptors are primarily expressed in the IPL, AMPA and KA receptors are expressed in both the OPL and the IPL. The presence of AMPA or KA receptors at OFF-bipolar cell dendrites in the OPL allows these cells to depolarize in response to photoreceptor glutamate release in the dark [99]

Metabotropic glutamate receptors

Bipolar cells, horizontal cells, retinal ganglion cells, and amacrine cells express metabotropic glutamate receptors (mGluRs) postsynaptically. Upon binding glutamate, mGluRs activate G-protein-dependent, second messenger cascades to convey the light signal. There are three groups of mGluRs, classified according to their secondary signaling cascades, pharmacology, and sequence homology. mGluR6, a group III mGluR, is expressed at ON-bipolar cell dendrites. Glutamate is normally released from photoreceptors in the dark, yet ON-bipolar cells depolarize in response to increments in light. Because glutamate binding of mGluR6 ultimately closes TRPM1 cation channels [100, 101], ON-bipolar cells can 'sign-invert' the photoreceptor signal, and thus hyperpolarize in the dark and depolarize in response to increased illumination [99]

Ribbon synapse

Ribbon synapses are excitatory synapses specialized for fast and tonic neurotransmitter release from photoreceptors and bipolar cells in the

**instead they
are tonically
active**

retina [1]. Unlike most neurons in the brain, photoreceptors and bipolar cells do not fire action potentials

encoding sensory information with graded changes in their membrane potential. Ribbon synapses feature specialized structures in order to continuously release neurotransmitter vesicles in response to changes in membrane potential. In fact, ribbon synapses are named for their most prominent presynaptic structure, the ribbon, which appears as a large, electron-dense bar or sheet at the active zone of the synapse with electron microscopy. Synaptic vesicles containing glutamate are tethered to ribbons, and transmitter is released upon calcium-dependent exocytosis. Another specialization is evident at photoreceptor synapses, where the dendritic processes of horizontal cells and ON bipolar cells invaginate the photoreceptor axonal terminal, placing postsynaptic glutamate receptors in close proximity to the presynaptic ribbon [102, 103]

References

1. Matthews G, Fuchs P. The diverse roles of ribbon synapses in sensory neurotransmission. *Nat Rev Neurosci.* 2010; 11:812–822. [PubMed: 21045860]
2. Maslim J, Stone J. Synaptogenesis in the retina of the cat. *Brain Res.* 1986; 373:35–48. [PubMed: 3719315]
3. Bodnarenko SR, Chalupa LM. Stratification of ON and OFF ganglion cell dendrites depends on glutamate-mediated afferent activity in the developing retina. *Nature.* 1993; 364:144–146. [PubMed: 8100613]
4. Bodnarenko SR, et al. Development and regulation of dendritic stratification in retinal ganglion cells by glutamate-mediated afferent activity. *J Neurosci.* 1995; 15:7037–7045. [PubMed: 7472459]
5. Kim IJ, et al. Laminar restriction of retinal ganglion cell dendrites and axons: subtype-specific developmental patterns revealed with transgenic markers. *J Neurosci.* 2010; 30:1452–1462. [PubMed: 20107072]
6. Mumm JS, et al. In vivo imaging reveals dendritic targeting of laminated afferents by zebrafish retinal ganglion cells. *Neuron.* 2006; 52:609–621. [PubMed: 17114046]
7. Morgan JL, et al. Axons and dendrites originate from neuroepithelial-like processes of retinal bipolar cells. *Nat Neurosci.* 2006; 9:85–92. [PubMed: 16341211]
8. Wassle H, et al. Cone contacts, mosaics, and territories of bipolar cells in the mouse retina. *J Neurosci.* 2009; 29:106–117. [PubMed: 19129389]
9. Helmstaedter M, et al. Connectomic reconstruction of the inner plexiform layer in the mouse retina. *Nature.* 2013; 500:168–174. [PubMed: 23925239]
10. Godinho L, et al. Targeting of amacrine cell neurites to appropriate synaptic laminae in the developing zebrafish retina. *Development.* 2005; 132:5069–5079. [PubMed: 16258076]
11. Johnson PT, et al. Rods and cones project to the inner plexiform layer during development. *J Comp Neurol.* 1999; 414:1–12. [PubMed: 10494074]
12. Sanes JR, Yamagata M. Many paths to synaptic specificity. *Annu Rev Cell Dev Biol.* 2009; 25:161–195. [PubMed: 19575668]
13. Yamagata M, et al. Sidekicks: synaptic adhesion molecules that promote lamina-specific connectivity in the retina. *Cell.* 2002; 110:649–660. [PubMed: 12230981]
14. Yamagata M, Sanes JR. Expanding the Ig superfamily code for laminar specificity in retina: expression and role of contactins. *J Neurosci.* 2012; 32:14402–14414. [PubMed: 23055510]

15. Yamagata M, Sanes JR. Dscam and Sidekick proteins direct lamina-specific synaptic connections in vertebrate retina. *Nature*. 2008; 451:465–469. [PubMed: 18216854]
16. Matsuoka RL, et al. Transmembrane semaphorin signalling controls laminar stratification in the mammalian retina. *Nature*. 2011; 470:259–263. [PubMed: 21270798]
17. Matsuoka RL, et al. Class 5 transmembrane semaphorins control selective Mammalian retinal lamination and function. *Neuron*. 2011; 71:460–473. [PubMed: 21835343]
18. Sun LO, et al. On and off retinal circuit assembly by divergent molecular mechanisms. *Science*. 2013; 342:1241974. [PubMed: 24179230]
19. Morgan JL, et al. Development of cell type-specific connectivity patterns of converging excitatory axons in the retina. *Neuron*. 2011; 71:1014–1021. [PubMed: 21943599]
20. Yoshimatsu T, et al. Transmission from the dominant input shapes stereotypic ration of photoreceptor inputs onto horizontal cells. *Nat Commun*. 2014; 5:3699. [PubMed: 24832361]
21. Dunn FA, Wong RO. Diverse strategies engaged in establishing stereotypic wiring patterns among neurons sharing a common input at the visual system's first synapse. *J Neurosci*. 2012; 32:10306–10317. [PubMed: 22836264]
22. Wei W, et al. Development of asymmetric inhibition underlying direction selectivity in the retina. *Nature*. 2011; 469:402–406. [PubMed: 21131947]
23. Yonehara K, et al. Spatially asymmetric reorganization of inhibition establishes a motion-sensitive circuit. *Nature*. 2011; 469:407–410. [PubMed: 21170022]
24. Huberman AD, et al. Mechanisms underlying development of visual maps and receptive fields. *Annu Rev Neurosci*. 2008; 31:479–509. [PubMed: 18558864]
25. Cang J, Feldheim DA. Developmental mechanisms of topographic map formation and alignment. *Annu Rev Neurosci*. 2013; 36:51–77. [PubMed: 23642132]
26. Xu HP, Tian N. Retinal ganglion cell dendrites undergo a visual activity-dependent redistribution after eye opening. *J Comp Neurol*. 2007; 503:244–259. [PubMed: 17492624]
27. Tian N, Copenhagen DR. Visual stimulation is required for refinement of ON and OFF pathways in postnatal retina. *Neuron*. 2003; 39:85–96. [PubMed: 12848934]
28. Xu HP, et al. The immune protein CD3zeta is required for normal development of neural circuits in the retina. *Neuron*. 2010; 65:503–515. [PubMed: 20188655]
29. Huh GS, et al. Functional requirement for class I MHC in CNS development and plasticity. *Science*. 2000; 290:2155–2159. [PubMed: 11118151]
30. Lee H, et al. Synapse elimination and learning rules co-regulated by MHC class I H2-D. *Nature*. 2014
31. Shatz CJ. MHC class I: an unexpected role in neuronal plasticity. *Neuron*. 2009; 64:40–45. [PubMed: 19840547]
32. Xu Y, et al. mGluR6 deletion renders the TRPM1 channel in retina inactive. *J Neurophysiol*. 2012; 107:948–957. [PubMed: 22131384]
33. Cao Y, et al. TRPM1 forms complexes with nyctalopin in vivo and accumulates in postsynaptic compartment of ON-bipolar neurons in mGluR6-dependent manner. *J Neurosci*. 2011; 31:11521–11526. [PubMed: 21832182]
34. Tagawa Y, et al. Immunohistological studies of metabotropic glutamate receptor subtype 6-deficient mice show no abnormality of retinal cell organization and ganglion cell maturation. *J Neurosci*. 1999; 19:2568–2579. [PubMed: 10087070]
35. Kerschensteiner D, et al. Neurotransmission selectively regulates synapse formation in parallel circuits in vivo. *Nature*. 2009; 460:1016–1020. [PubMed: 19693082]
36. Okawa H, et al. Interplay of cell-autonomous and nonautonomous mechanisms tailors synaptic connectivity of converging axons in vivo. *Neuron*. 2014; 82:125–137. [PubMed: 24698272]
37. Bleckert A, Wong RO. Identifying roles for neurotransmission in circuit assembly: insights gained from multiple model systems and experimental approaches. *Bioessays*. 2011; 33:61–72. [PubMed: 21110347]
38. Dunn FA, et al. Sensory experience shapes the development of the visual system's first synapse. *Neuron*. 2013; 80:1159–1166. [PubMed: 24314727]

39. Dick O, et al. The presynaptic active zone protein bassoon is essential for photoreceptor ribbon synapse formation in the retina. *Neuron*. 2003; 37:775–786. [PubMed: 12628168]
40. Regus-Leidig H, et al. Absence of functional active zone protein Bassoon affects assembly and transport of ribbon precursors during early steps of photoreceptor synaptogenesis. *Eur J Cell Biol*. 2010; 89:468–475. [PubMed: 20188438]
41. Raven MA, et al. Early afferent signaling in the outer plexiform layer regulates development of horizontal cell morphology. *J Comp Neurol*. 2008; 506:745–758. [PubMed: 18076080]
42. Bayley PR, Morgans CW. Rod bipolar cells and horizontal cells form displaced synaptic contacts with rods in the outer nuclear layer of the nob2 retina. *J Comp Neurol*. 2007; 500:286–298. [PubMed: 17111373]
43. Zabouri N, Haverkamp S. Calcium channel-dependent molecular maturation of photoreceptor synapses. *PLoS One*. 2013; 8:e63853. [PubMed: 23675510]
44. Van Epps HA, et al. The zebrafish nrc mutant reveals a role for the polyphosphoinositide phosphatase synaptojanin 1 in cone photoreceptor ribbon anchoring. *J Neurosci*. 2004; 24:8641–8650. [PubMed: 15470129]
45. Allwardt BA, et al. Synapse formation is arrested in retinal photoreceptors of the zebrafish nrc mutant. *J Neurosci*. 2001; 21:2330–2342. [PubMed: 11264308]
46. Jia S, et al. Zebrafish Cacna1fa is required for cone photoreceptor function and synaptic ribbon formation. *Hum Mol Genet*. 2014
47. Sato S, et al. Pikachurin, a dystroglycan ligand, is essential for photoreceptor ribbon synapse formation. *Nat Neurosci*. 2008; 11:923–931. [PubMed: 18641643]
48. Stafford BK, et al. Developmental changes in NMDA receptor subunit composition at ON and OFF bipolar cell synapses onto direction-selective retinal ganglion cells. *J Neurosci*. 2014; 34:1942–1948. [PubMed: 24478373]
49. Hartong DT, et al. Retinitis pigmentosa. *Lancet*. 2006; 368:1795–1809. [PubMed: 17113430]
50. Jones BW, et al. Retinal remodeling. *Jpn J Ophthalmol*. 2012; 56:289–306. [PubMed: 22644448]
51. Puthussery T, Taylor WR. Functional changes in inner retinal neurons in animal models of photoreceptor degeneration. *Adv Exp Med Biol*. 2010; 664:525–532. [PubMed: 20238055]
52. Calkins DJ. Critical pathogenic events underlying progression of neurodegeneration in glaucoma. *Prog Retin Eye Res*. 2012; 31:702–719. [PubMed: 22871543]
53. Chang B, et al. Retinal degeneration mutants in the mouse. *Vision Res*. 2002; 42:517–525. [PubMed: 11853768]
54. Phillips MJ, et al. Progression of neuronal and synaptic remodeling in the rd10 mouse model of retinitis pigmentosa. *J Comp Neurol*. 2010; 518:2071–2089. [PubMed: 20394059]
55. Chang B, et al. Two mouse retinal degenerations caused by missense mutations in the beta-subunit of rod cGMP phosphodiesterase gene. *Vision Res*. 2007; 47:624–633. [PubMed: 17267005]
56. Gargini C, et al. Retinal organization in the retinal degeneration 10 (rd10) mutant mouse: a morphological and ERG study. *J Comp Neurol*. 2007; 500:222–238. [PubMed: 17111372]
57. Barhoum R, et al. Functional and structural modifications during retinal degeneration in the rd10 mouse. *Neuroscience*. 2008; 155:698–713. [PubMed: 18639614]
58. Puthussery T, et al. Differential loss and preservation of glutamate receptor function in bipolar cells in the rd10 mouse model of retinitis pigmentosa. *Eur J Neurosci*. 2009; 29:1533–1542. [PubMed: 19385989]
59. Keeley PW, Reese BE. Role of afferents in the differentiation of bipolar cells in the mouse retina. *J Neurosci*. 2010; 30:1677–1685. [PubMed: 20130177]
60. Della Santina L, et al. Differential progression of structural and functional alterations in distinct retinal ganglion cell types in a mouse model of glaucoma. *J Neurosci*. 2013; 33:17444–17457. [PubMed: 24174678]
61. Weber AJ, et al. Morphology of single ganglion cells in the glaucomatous primate retina. *Invest Ophthalmol Vis Sci*. 1998; 39:2304–2320. [PubMed: 9804139]
62. Jakobs TC, et al. Retinal ganglion cell degeneration is topological but not cell type specific in DBA/2J mice. *J Cell Biol*. 2005; 171:313–325. [PubMed: 16247030]

63. Feng L, et al. Sustained ocular hypertension induces dendritic degeneration of mouse retinal ganglion cells that depends on cell type and location. *Invest Ophthalmol Vis Sci.* 2013; 54:1106–1117. [PubMed: 23322576]
64. Peng YW, et al. Ectopic synaptogenesis in the mammalian retina caused by rod photoreceptor-specific mutations. *Nat Neurosci.* 2000; 3:1121–1127. [PubMed: 11036269]
65. Cuenca N, et al. Regressive and reactive changes in the connectivity patterns of rod and cone pathways of P23H transgenic rat retina. *Neuroscience.* 2004; 127:301–317. [PubMed: 15262321]
66. Strettoi E, et al. Recruitment of the rod pathway by cones in the absence of rods. *J Neurosci.* 2004; 24:7576–7582. [PubMed: 15329405]
67. Haverkamp S, et al. Synaptic plasticity in CNGA3(-/-) mice: cone bipolar cells react on the missing cone input and form ectopic synapses with rods. *J Neurosci.* 2006; 26:5248–5255. [PubMed: 16687517]
68. Fariss RN, et al. Abnormalities in rod photoreceptors, amacrine cells, and horizontal cells in human retinas with retinitis pigmentosa. *Am J Ophthalmol.* 2000; 129:215–223. [PubMed: 10682975]
69. Marc RE, et al. Neural remodeling in retinal degeneration. *Prog Retin Eye Res.* 2003; 22:607–655. [PubMed: 12892644]
70. Strettoi E, Pignatelli V. Modifications of retinal neurons in a mouse model of retinitis pigmentosa. *Proc Natl Acad Sci U S A.* 2000; 97:11020–11025. [PubMed: 10995468]
71. Strettoi E, et al. Remodeling of second-order neurons in the retina of rd/rd mutant mice. *Vision Res.* 2003; 43:867–877. [PubMed: 12668056]
72. Strettoi E, et al. Morphological and functional abnormalities in the inner retina of the rd/rd mouse. *J Neurosci.* 2002; 22:5492–5504. [PubMed: 12097501]
73. Chang B, et al. The nob2 mouse, a null mutation in *Cacna1f*: anatomical and functional abnormalities in the outer retina and their consequences on ganglion cell visual responses. *Vis Neurosci.* 2006; 23:11–24. [PubMed: 16597347]
74. Michalakakis S, et al. Characterization of neurite outgrowth and ectopic synaptogenesis in response to photoreceptor dysfunction. *Cell Mol Life Sci.* 2013; 70:1831–1847. [PubMed: 23269435]
75. Sonntag S, et al. Ablation of retinal horizontal cells from adult mice leads to rod degeneration and remodeling in the outer retina. *J Neurosci.* 2012; 32:10713–10724. [PubMed: 22855819]
76. Keeley PW, et al. Development and plasticity of outer retinal circuitry following genetic removal of horizontal cells. *J Neurosci.* 2013; 33:17847–17862. [PubMed: 24198374]
77. Dhingra A, et al. The light response of ON bipolar neurons requires G[alpha]o. *J Neurosci.* 2000; 20:9053–9058. [PubMed: 11124982]
78. Ball SL, et al. Immunohistochemical analysis of the outer plexiform layer in the nob mouse shows no abnormalities. *Vis Neurosci.* 2003; 20:267–272. [PubMed: 14570248]
79. Cao Y, et al. Retina-specific GTPase accelerator RGS11/G beta 5S/R9AP is a constitutive heterotrimer selectively targeted to mGluR6 in ON-bipolar neurons. *J Neurosci.* 2009; 29:9301–9313. [PubMed: 19625520]
80. Regus-Leidig H, et al. Photoreceptor degeneration in two mouse models for congenital stationary night blindness type 2. *PLoS One.* 2014; 9:e86769. [PubMed: 24466230]
81. Matsuoka RL, et al. Guidance-cue control of horizontal cell morphology, lamination, and synapse formation in the mammalian outer retina. *J Neurosci.* 2012; 32:6859–6868. [PubMed: 22593055]
82. Soto F, et al. NGL-2 regulates pathway-specific neurite growth and lamination, synapse formation, and signal transmission in the retina. *J Neurosci.* 2013; 33:11949–11959. [PubMed: 23864682]
83. Chua J, et al. Functional remodeling of glutamate receptors by inner retinal neurons occurs from an early stage of retinal degeneration. *J Comp Neurol.* 2009; 514:473–491. [PubMed: 19350664]
84. Marc RE, et al. Neural reprogramming in retinal degeneration. *Invest Ophthalmol Vis Sci.* 2007; 48:3364–3371. [PubMed: 17591910]
85. Jones BW, et al. Retinal remodeling in the Tg P347L rabbit, a large-eye model of retinal degeneration. *J Comp Neurol.* 2011; 519:2713–2733. [PubMed: 21681749]
86. Cuenca N, et al. Changes in the inner and outer retinal layers after acute increase of the intraocular pressure in adult albino Swiss mice. *Exp Eye Res.* 2010; 91:273–285. [PubMed: 20650699]

87. Fuchs M, et al. Rod photoreceptor ribbon synapses in DBA/2J mice show progressive age-related structural changes. *PLoS One*. 2012; 7:e44645. [PubMed: 22957094]
88. Chen M, et al. Development and degeneration of cone bipolar cells are independent of cone photoreceptors in a mouse model of retinitis pigmentosa. *PLoS One*. 2012; 7:e44036. [PubMed: 22952865]
89. O'Brien EE, et al. The effect of photoreceptor degeneration on ganglion cell morphology. *J Comp Neurol*. 2014; 522:1155–1170. [PubMed: 24519018]
90. Reynolds J, Lamba DA. Human embryonic stem cell applications for retinal degenerations. *Exp Eye Res*. 2013
91. Wright LS, et al. Induced pluripotent stem cells as custom therapeutics for retinal repair: Progress and rationale. *Exp Eye Res*. 2014
92. Pearson RA. Advances in repairing the degenerate retina by rod photoreceptor transplantation. *Biotechnol Adv*. 2014; 32:485–491. [PubMed: 24412415]
93. Gallina D, et al. A comparative analysis of Muller glia-mediated regeneration in the vertebrate retina. *Exp Eye Res*. 2013
94. Fuhrmann S, et al. Retinal pigment epithelium development, plasticity, and tissue homeostasis. *Exp Eye Res*. 2013
95. Hertz J, et al. Survival and Integration of Developing and Progenitor-Derived Retinal Ganglion Cells Following Transplantation. *Cell Transplant*. 2013
96. Deans MR, et al. Control of neuronal morphology by the atypical cadherin Fat3. *Neuron*. 2011; 71:820–832. [PubMed: 21903076]
97. Jan YN, Jan LY. Branching out: mechanisms of dendritic arborization. *Nat Rev Neurosci*. 2010; 11:316–328. [PubMed: 20404840]
98. Gorsuch RA, Hyde DR. Regulation of Muller glial dependent neuronal regeneration in the damaged adult zebrafish retina. *Exp Eye Res*. 2013
99. Yoshimatsu, T., et al. Circuit Assembly in the Developing Vertebrate Retina. In: Rubenstein, JLR.; Rakic, P., editors. *Cellular Migration and Formation of Neuronal Connections*. Academic Press; 2013. p. 687-711.
100. Morgans CW, et al. TRPM1 is required for the depolarizing light response in retinal ON-bipolar cells. *Proc Natl Acad Sci U S A*. 2009; 106:19174–19178. [PubMed: 19861548]
101. Koike C, et al. TRPM1 is a component of the retinal ON bipolar cell transduction channel in the mGluR6 cascade. *Proc Natl Acad Sci U S A*. 2010; 107:332–337. [PubMed: 19966281]
102. Schmitz F. The making of synaptic ribbons: how they are built and what they do. *Neuroscientist*. 2009; 15:611–624. [PubMed: 19700740]
103. Regus-Leidig H, Brandstatter JH. Structure and function of a complex sensory synapse. *Acta Physiol (Oxf)*. 2012; 204:479–486. [PubMed: 21880116]
104. Wassle H. Parallel processing in the mammalian retina. *Nat Rev Neurosci*. 2004; 5:747–757. [PubMed: 15378035]
105. Bleckert A, et al. Spatial relationships between GABAergic and glutamatergic synapses on the dendrites of distinct types of mouse retinal ganglion cells across development. *PLoS One*. 2013; 8:e69612. [PubMed: 23922756]
106. Doering CJ, et al. Modified Ca(v)1.4 expression in the *Cacna1f(nob2)* mouse due to alternative splicing of an ETn inserted in exon 2. *PLoS One*. 2008; 3:e2538. [PubMed: 18596967]
107. Dhingra A, et al. Gbeta3 is required for normal light ON responses and synaptic maintenance. *J Neurosci*. 2012; 32:11343–11355. [PubMed: 22895717]

Highlights

- Synaptic refinement shapes the stereotypic connectivity of many, but not all, retinal circuits
- Novel connections can form during retinal degeneration
- Miswiring in circuit disassembly may constrain circuit repair

Figure 1. Organization of retinal structure, connectivity and synapses

(A) Cross-section of the adult mouse retina (cover image from [7]).

Cone photoreceptors (purple), horizontal cells (orange), bipolar cells (green), amacrine cells and retinal ganglion cells are labeled (red). Outer nuclear layer (ONL); inner nuclear layer (INL); retinal ganglion cell layer (GCL). Outer plexiform layer (OPL); inner plexiform layer (IPL).

(B) Schematic showing major retinal pathways.

Rod and cone photoreceptors detect changes in illumination, with rods functioning at low light levels, and cones at high light levels. Photoreceptor signals are conveyed by bipolar

cells to retinal ganglion cells (RGCs). Cone bipolar cells (CBC) that largely contact cone photoreceptors are grouped into two major subclasses. Light increments depolarize ON-bipolar cells and hyperpolarize OFF-bipolar cells. ON and OFF synaptic connections are organized into separate laminae within the IPL. Horizontal cells (HC) and amacrine cells (AC) modulate information flow in the outer and inner retina, respectively. Rod bipolar cells (RBCs) predominantly convey rod input, and contact AII amacrine cells (AII AC) that inhibit transmission from OFF-CBCs.

(C) Subcellular organization of retinal synapses.

Schematic of a cone photoreceptor ribbon synapse (left). ON-CBCs and HCs invaginate the cone pedicle at sites apposed to a ribbon structure with tethered synaptic vesicles. OFF-CBCs make basal contacts with the cone pedicle [104]. mGluR6, metabotropic glutamate receptors on ON BCs; iGluR, ionotropic glutamate receptor on OFF-CBCs. Electron micrograph of a zebrafish cone pedicle at 5 days post-fertilization (right, from Yoshimatsu et al. [20]). HC and CBC processes are pseudocolored as in the schematic.

(D) Schematic (left) and ultrastructure (right) of synapses in the IPL of an adult mouse retina. Shown here are BC and AC synapses. A ribbon synapse (small arrow) between a CBC and an RGC is apparent. The AC here provides feed-forward (FF, asterisk) inhibition onto the RGC and feedback (FB, large arrow) inhibition onto the BC on the right.

Micrograph provided by A. Bleckert [105].

Figure 2. Strategies effecting RGC dendritic lamination and synaptic partner choice

At least four cellular strategies are employed to generate dendritic lamination patterns of RGCs and their connectivity. Distinct RGC types engage dendritic pruning to different extents (1,2,3) to achieve their final stratification pattern. Addition of arbors (4), instead of pruning, also occurs. Retinal ganglion cell, RGC; cone bipolar cells, C; rod bipolar cells, R. Orange, ON bipolar cell; Blue, OFF bipolar cell; Gray, rod bipolar cell.

Figure 3. Remodeling and perturbations to retinal excitatory synapses

(A) Mouse mutants in which ribbon arrangements are altered in cone photoreceptor terminals. Bassoon, cytomatrix protein [39, 40]; Cacna1f, voltage-gated calcium channel [41, 43]; Pikachurin, dystroglycan ligand [47]; WT, wildtype.

(B) Morphological defects in rod photoreceptor terminals. *nob2*, partial loss-of-function mutation in *cacna1f* [42, 106], *nob4*, point mutation in mGluR6 [79]; *NGL-2*, netrin-G ligand 2 [82], *Sema6*, semaphorin 6A; *PlexA4*, Plexin A4 [81], *Gnb3*, G-protein beta subunit 3 [107].

Figure 4. Structural and functional rewiring in disease

(A) In healthy retina, rod bipolar cells (RBCs) synapse primarily with rod photoreceptors, localizing mGluR6 to invaginating dendritic tips apposed to synaptic ribbons. (Left) Sprouting of RBC dendrites into the ONL in rod transmission-defective mutants [43,73]. (Right) Rod degeneration in the *rd10* model of Retinitis Pigmentosa (RP) leads to retraction of RBC dendrites [54, 56–58], and a simultaneous loss or redistribution of mGluR6 to the cell body and axon [56, 57]. RBC dendritic retraction may precede the formation of synapses with novel partners, such as cone photoreceptors [54, 56, 58].

(B) Changes in receptor localization and expression at synaptic contacts between bipolar cells and cone photoreceptors in disease models (*rd1* and *rd10*) [56–58, 82] and in human RP[83–85].

Figure I. Phenotypes of mouse mutants with synapse lamination defects

In wild type retina (WT), neurites of tyrosine hydroxylase-expressing amacrine cells (TH) arborize in the OFF sublamina of the IPL, where they make synapses with the dendrites of M1 type melanopsin-expressing retinal ganglion cells (M1 RGC). AII amacrine cells (AII) and rod bipolar cells (RBC) form synaptic contacts in the inner tier of the ON sublamina. In *Sema5A/5B* double mutant mice (*Sema5A/5B* dKO), neurites of TH expressing amacrine cells and AII amacrine cells arborize erroneously in the inner nuclear layer (INL). The dendrites of M1 RGCs also extend into the INL and associate with TH positive neurites. In the *Sema6A* mutant (*Sema6A* KO), TH amacrine cell processes invade the ON sublamina, where they associate with aberrant M1 RGC dendrites. Horizontal cell axons also overshoot into the outer nuclear layer (ONL). It remains to be seen whether AII amacrine cells are affected in the *Sema6A* mutant. In the *Fat3* mutant (*Fat3* KO), TH amacrine cells project extra processes toward the outer misplaced plexiform layer (OMPL) in the INL. AII amacrine cells send extra neurites to the OMPL and the OPL. Although to date there is no data to show whether there is an effect on M1 RGCs, synaptic coupling between AII amacrine cells and rod bipolar cells is maintained but mislocalized to the inner misplaced plexiform layer (IMPL) beneath the retinal ganglion cell layer (GCL).