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not extend to seeing and escaping danger.

DECLARATION OF INTERESTS

The authors declare no competing interests.

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Neuroscience: Visual restoration with optogenetics

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Treating photoreceptor degenerative diseases is an exciting application of optogenetic technologies. However, there are significant challenges, such as producing normal visual signaling as the retina rewires in response to photoreceptor death. However, a new study shows remarkable functional stability in retinal circuits that can be engaged by optogenetics following photoreceptor loss.

Photoreceptor degenerative diseases, such as age-related macular degeneration and retinitis pigmentosa, result in the massive loss of light-detecting cells in the retina and ultimately lead to blindness. Currently, there are no cures for these diseases, but many treatments are being developed[1]. These include implanted electrical prosthetics to electrically stimulate retinal neurons, replacing lost photoreceptors with precursor cells, and gene therapies that aim to insert a healthy copy of the mutated gene that is causing the degeneration. A fourth approach, examined by Rodgers, Hughes *et al.*[2] in this issue of *Current Biology*, aims to express an exogenous light-sensitive molecule, ReaChR[3], in downstream retinal neurons, effectively converting them into photoreceptors[4,5]. Each of these approaches presents a range of advantages and challenges toward implementation, but all are exciting avenues for curing blindness.

One challenge faced by all attempts at restoring vision is that neural circuits in the retina rewire as photoreceptors die[6]. For example, retinal bipolar cells and horizontal cells, which synapse with photoreceptors, initially extend dendrites to find new synapses as photoreceptor inputs are lost[7]. However, if there are no photoreceptors nearby, the cells retract their dendrites and downregulate synaptic proteins. Downstream amacrine cells and retinal ganglion cells also undergo synaptic plasticity in response to bipolar and horizontal cell rewiring[8]. Finally, Muller glia 'seal' the neural retina from the pigment epithelium and choroid[9]. Coinciding with these anatomical changes are changes in physiology. For example, retinal ganglion cells exhibit aberrant oscillatory spiking in some animal models[10–12]. This aberrant activity could limit the fidelity of the signals transmitted from the retina to the rest of the brain even after therapy. Cumulatively, these reports of impaired structure and function suggest rewiring is maladaptive and may be an impediment to restoring vision.

However, other studies suggest retinal rewiring supports visual signaling in the face of degeneration[13]. For example, rewiring by bipolar cells can serve to maintain visual signaling rather than impair intact circuits[14]. Furthermore, following partial rod ablation, retinal ganglion cell activity remains surprisingly robust, indicating retinal circuitry is capable of amplifying reduced input to maintain vision[15]. Robust cone-mediated signals also persist for much longer than anatomy would suggest in degenerating retinas[16]. However, all these studies are in the context of retinas that have some number of remaining photoreceptors.

Does the retina retain circuitry that could be reactivated once all or nearly all the photoreceptors have been lost?

The results from Rodgers, Hughes *et al.*[2] strongly suggest 'yes'! The authors approached this question using a degenerating mouse line that expresses a variant of ChannelRhodopsin2 called ReaChR in ON bipolar cells[3]. This effectively turned the ON bipolar cells into photoreceptors, causing these neurons to depolarize in response to light. Note, this is approximately how these neurons would respond to an increase in light. The idea was as follows: if, on the one hand, the retina has undergone substantial rewiring that interferes with visual processing, then stimulating the ON bipolar cells following photoreceptor death would lead to an aberrant set of visual responses among the retinal ganglion cells and in retino-recipient areas of the brain, such as the visual thalamus. On the other hand, if retinal circuits were largely normal, then signals among retinal ganglion cells and retino-recipient areas should also be relatively normal.

The authors compared the distribution of response properties observed in normal mice, both among retinal ganglion cells and in the visual thalamus, with those measured in mice that had no remaining rods or cones, but were expressing ReaChR in the ON bipolar cells. They found remarkably similar distributions of different kinds of visual responses between the two cohorts. Even retinal ganglion cells that respond only to particular directions of motion — a relatively sophisticated computation performed by retinal circuits — remained in the mice lacking rods and cones. The largest divergence between the normal mice and those lacking rods and cones was a reduced number of 'OFF' responses, which was unsurprising given that 'OFF' responses are initiated by OFF bipolar cells and these were not directly activated by light in the ReaChR-expressing mice. Nevertheless, some OFF responses were present at the level of retinal ganglion cells, probably via the phenomenon of crossover inhibition, which is present in healthy retinal circuits[17]. Another interesting difference between the two cohorts was that the visual neurons in the ReaChR mice could respond to higher temporal frequency stimuli, likely because ReaChR mediates much more rapid phototransduction than the G-protein coupled receptor cascades engaged by endogenous opsins in the photoreceptors. Overall, the authors demonstrate that visual responses generated from exogenous opsins in ON bipolar cells had similar properties to those in mice with normal vision. Thus, retinal rewiring does not appear to strongly perturb the gross features of retinal signaling to the rest of the brain, even after the photoreceptors have died. This observation provides great hope for bringing treatments for blindness developed in the lab to humans.

Given that many people are not diagnosed with retinal degeneration until they have lost a large fraction of photoreceptors, it is essential to create therapies specific to late-stage degenerations. Optogenetic therapy entails creating new photosensitive cells out of existing retinal cells, and is thus an attractive approach. Most optogenetic therapies to date have delivered opsins to retinal ganglion cells, primarily because these cells are the most accessible to viral therapy in humans [5]. However, introducing opsins earlier in the retinal circuitry may support more normal and diverse visual signaling to the brain, as evidenced in the study from Rodgers, Hughes *et al.*[2]. Consistent with these results, several studies that used adeno-associated viruses to introduce opsins into mouse ON bipolar cells show promising recovery of at least some visual signaling[4,18–20]. Thus, future studies should compare optogenetic-mediated vision from bipolar cells versus retinal ganglion cells.

Another important direction for future work is to determine the extent to which optogenetic therapies allow for vision in more natural contexts. Natural scenes engage nonlinearities in retinal processing that are not often engaged by artificial stimuli such as spots of light, flashes, and sinusoidal gratings. As a result, natural scene stimuli and natural behavioral tasks are becoming more common to assay rodent vision. Such studies could prove decisive in determining which aspects of retinal rewiring are maladaptive and which facilitate robust visual signaling in response to cell death.

DECLARATION OF INTERESTS

The authors declare no competing interests.

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Eukaryotic evolution: Deep phylogeny does not imply morphological novelty

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Eukaryotic diversity is often depicted as a molecular phylogenetic tree consisting of a few supergroups that originated over a billion years ago. A new study reveals an ancient group of tiny phagotrophic flagellates that reinforces inferences about early evolutionary history.

Microbial organisms have dominated the planet in both abundance and phylogenetic diversity for over three billion years, yet the novel traits of many of these lineages remain poorly understood or completely unknown. It is well known, however, that some microbes are more complex than others at both morphological and genetic levels, reflecting diverse modes of nutrition, ecological roles, and evolutionary histories. For instance, the origin of eukaryotes about a billion years ago involved several key innovations that increased cellular complexity, including the emergence of a cytoskeletal system that facilitates locomotion, reproduction, and the ingestion of bacterial prey cells through a process called 'phagotrophy'[1]. Although several lineages of modern phagotrophic eukaryotes have retained their appetite for bacteria, some phagotrophic eukaryotes eat other microbial eukaryotes instead. Phagotrophic modes of nutrition are inferred to reflect the early evolutionary history of eukaryotes, which ultimately set the stage for the astounding array of biological novelty found within the known supergroups, such as the independent origins of multicellularity, parasitism, secreted cell coverings, intracellular armor, extrusomes, and photosynthesis via endosymbiosis. A new study by Tikhonenkov *et al.*[2] sheds additional light onto the earliest stages of eukaryotic evolution through the cultivation and characterization of several new strains of tiny phagotrophic flagellates (< 10 μm) that not only have retained many ancestral traits, but collectively form a new group of eukaryotes, called the Provora.

The first known member of the Provora, *Ancoracysta twista*, was described in 2017 by Janouškovec *et al.*[3] and, at the time, was considered an orphan lineage within the tree of eukaryotes, because it did not nest within any of the known supergroups[4]. Advances in phylogenomics and phylotranscriptomics have led to the discoveries of several other orphan lineages of tiny, morphologically streamlined phagotrophic flagellates[4–8]. The newly reported Provora now contains seven species that were collected from diverse and distant geographical locations, including the Caribbean Sea, Black Sea, Red Sea, Pacific Ocean, and Atlantic Ocean. The global distribution of this new group is bolstered by re-analyses of previously published environmental rDNA sequence surveys of eukaryotic diversity[2]. These analyses also suggest that members of the Provora might be numerically rare in specific locations despite their cosmopolitan distributions, making single-cell isolation and cultivation efforts particularly important.

However, there are several reasons beyond rarity that would explain why it is difficult to find tiny phagotrophic flagellates with global distributions, the most important of which are that sampling tiny fast-moving cells from a specific spot within a vast continuous ocean is essentially random and our overall knowledge of marine microbial diversity is