# “Recycling for sight”

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‘subject strapline’: Retinal biology

‘standfirst’: Vision requires the continuous recycling of photobleached pigments. A non-canonical type of autophagy in retinal pigment epithelial cells participates in this process.

Vision begins with the absorption of light by light-sensitive photoreceptors in the retina. After the arrival of photons, chromophores present in these cells undergo conformational changes and trigger the phototransduction cascade, which converts light into electrical impulses that travel to the brain and are transformed into the images we see. To sustain vision, the chromophore 11-*cis*-retinal, must be recycled back into their original conformation through a process known as the visual cycle. This cycle involves tight regulation of the interaction between photoreceptors and the cells of the adjacent retinal pigment epithelium (RPE), which nourish photoreceptors and improve the quality of the optical system of the eye by absorbing scattered light. A second key role of RPE cells is the degradation and recycling of the outer segments of photoreceptor cells, which are damaged by the impact of millions of photons per day. Each morning, these outer segments are engulfed (phagocytosed) by the highly polarized cells of the RPE and degraded inside acidic organelles called lysosomes. Whereas this phagocytic process has been known for decades, the intimate molecular and cellular mechanisms of chromophore and photoreceptor recycling have posed important challenges. In a recent issue of Cell, Kim *et al.*[1] demonstrate for the first time that, selected proteins implicated in autophagy, - a “self-eating” pathway that recycles cellular components after degrading them inside lysosomes- are necessary for the degradation of photoreceptor outer segments. They also show that vision is impaired and chromophore levels are reduced in mice that specifically lack the autophagy regulator Atg5 in the RPE. More

importantly, retinoid supplementation in these mice restored vision, revealing a new and unexpected link between autophagic degradation and chromophore regeneration in sustaining the visual cycle.

The study found that phagocytosis of photoreceptor outer segments coincided with an increase in the levels of the autophagy marker LC3-II, and involved a non-canonical autophagy pathway called LC3-associated phagocytosis (LAP). This process required canonical autophagy regulators such as Atg5 (Figure 1). However, as previously described for other processes involving LAP (*e.g.,* the elimination of intracellular pathogens and apoptotic cells)[2], it was independent of the proteins of the autophagy pre-initiation complex. Given that the vesicle to be degraded (phagosome) is already formed in LAP, it is perhaps unsurprising that this process is independent of the autophagy pre-initiation complex. This contrasts with canonical autophagy, which requires the formation of the autophagosome to sequester cytoplasmic components[3].

The classical route of phagosome maturation involves their acidification and acquisition of degradative enzymes to break down the ingested material. Why then are autophagy proteins required for LAP? The current hypothesis is that the translocation of autophagy proteins to the phagosomal membrane allows for their rapid fusion with lysosomes enhancing degradation[4]. In support of this view, Kim and coworkers observed no defects in phagocytic uptake in Atg5-deficient RPE cells. However, the phagosomes in these cells failed to migrate towards the basal side of the epithelium, where fusion with lysosomes occurs. Furthermore, protease maturation was impaired and the expression of lysosomal membrane glycoproteins was reduced in these phagosomes. It thus appears that this non-canonical form of autophagy promotes the trafficking of phagosomes and their fusion with lysosomes, ensuring rapid recycling of photoreceptor outer segments.

A second key finding of the study reveals a role for autophagy proteins in the visual cycle, a transcellular process though which RPE cells maintain the supply of chromophores that are required for the regeneration of visual pigment in photoreceptors (Figure 1). Upon light absorption in photoreceptors, the chromophore 11-*cis*-retinal is isomerized into all-*trans*-retinal. To regenerate 11-*cis*-retinal, all-*trans*-retinal is released from opsin and reduced to all-*trans*-retinol (vitamin A), which diffuses to the intercellular space and enters inside the adjacent RPE cells where it is transformed back into 11-*cis*-retinal[5]. Vitamin A is also provided by the blood supply and through the phagocytosis of the photoreceptor outer segment by the RPE[6]. Kim and colleagues have

demonstrated for the first time that autophagy proteins are required to recycle the 11-*cis*-retinal after phagocytosis of photoreceptor outer segments. Although many aspects of this process remain unclear, it may represent a highly efficient mechanism to recover chromophores from discarded photoreceptor outer segments. Each day, 10% of the total ocular retinoid pool passes through the RPE cell phagolysosomal system during the photoreceptor outer segment shedding. This study suggests that at least a fraction this 11-*cis*-retinal can be recovered and used to regenerate visual pigment without the involvement of intercellular transport systems or enzymes of the visual cycle, which are often mutated in retinal diseases[6].

Interestingly, retinal aging is associated with decreased autophagic activity and a parallel reduction in night vision, a phenotype that is mimicked by Atg5 deletion in retinal precursors[7]. In the lens, defects in autophagy protein function result in age-related cataract[8]. Moreover, retinal ganglion cells lacking Atg4 and Atg5 are more sensitive to optic nerve damage[9]. A common feature of these autophagy-impaired mutants is the accumulation of toxic products suggesting a defect in intracellular quality control. The accumulation of lipofuscin, an autofluorescent cellular waste product derived from the incomplete digestion of outer segments, is also frequently observed in human retinal diseases, such as age dependent macular degeneration. This disease is caused by a primary malfunction of the retinal pigment epithelium, which leads to photoreceptor death and subsequent blindness. In their Atg5-deficient mutants, Kim *et al*. did not report an increase in lipofuscin levels and photoreceptor numbers remain normal, even after seven months. Moreover, they successfully reversed the effects of Atg5 deletion by retinoid supplementation, demonstrating that autophagy downregulation primarily results in alterations in the visual cycle and not defects in intracellular quality control, as previously thought. Vitamin A administration and retinoid supplementation have also been used to treat some retinopathies associated with visual cycle defects and aging. Interestingly, retinoid derivatives increase the activity of chaperone-mediated autophagy[10], another lysosomal pathway that is upregulated in the retina as a compensatory response to age-associated decreases in macroautophagy[7]. Taken together, these findings suggest that retinoids hold significant potential for the treatment of retinal diseases and illuminate the role of autophagy proteins in vision fitness.

**Figure 1:** Light enters the eye and strikes the mouse retina (a), where it photoactivates pigments in the membranes of the outer segments of photoreceptors. This induces the release of the bleached pigment (opsin) and a molecule of all-*trans*-retinal that is next converted into vitamin A (all-*trans*-retinol), which is recycled back into the chromophore 11-*cis*-retinal via the visual cycle (yellow arrows). The binding of 11-*cis*-retinal to the opsin restores the photoactivable pigment rhodopsin that can react again with light and start the phototransduction cascade (b). The impact of light damages the membranes of rod photoreceptor outer segments, which are subsequently phagocytosed by cells of the retinal-pigment epithelium (RPE) (c). After the phagosome is formed, a non-canonical autophagy pathway involving Atg5 and LC3 triggers its fusion with the lysosome, resulting in degradation of the outer segments. The degradation products exit the autophagolysosome and are recycled to sustain the visual cycle.


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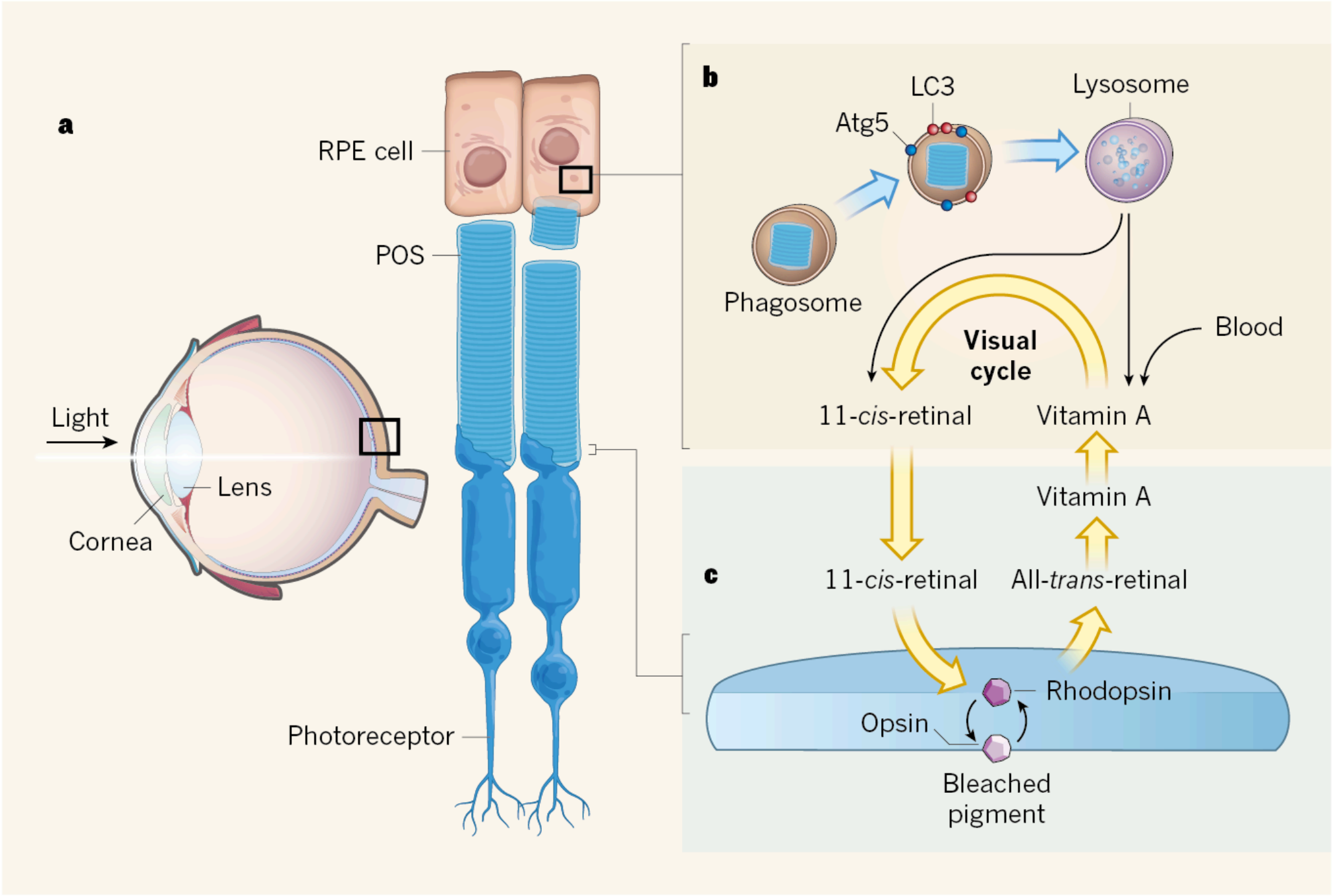
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RPE cell
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