

NEUROPROTECTION AND GLAUCOMA: OLD CHALLENGES, NEW SOLUTIONS

New drug targets and approaches to designing clinical trials hold promise for the development of neuroprotective therapies for glaucoma.



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Glaucoma is a progressive optic neuropathy characterized by a loss of retinal ganglion cells (RGCs), leading to visual field defects and, ultimately, blindness. The pathophysiology of this disease remains incompletely understood. Although IOP is considered to be the most important and only modifiable risk factor in the development of glaucoma,¹⁻³ RGC loss continues in some patients despite lowering of the IOP, suggesting that other risk factors may contribute to neuronal loss in glaucoma.^{4,5} The mechanism of RGC apoptosis is likely multifactorial, with contributors including deprivation of neurotrophic factors, mitochondrial dysfunction, glial activation, excitotoxicity, failure of axonal transport, and oxidative stress (Figure 1).^{6,7}

Neuroprotection in glaucoma is defined as an intervention that targets the molecular mechanisms causing RGC death and thereby enhances the survival of RGCs independent of IOP control. There is great interest in finding therapies that can offer neuroprotection in glaucoma. The course of research and development in this area

has been rocky, but new approaches hold promise.

HISTORICAL APPROACHES

Investigators have tested many molecules for their neuroprotective effects in glaucoma, but success

in their translation to clinical use has been limited. A case in point is memantine, a noncompetitive N-methyl-D-aspartate subtype of glutamate receptor antagonist. Already in use for treating Alzheimer disease, memantine showed early

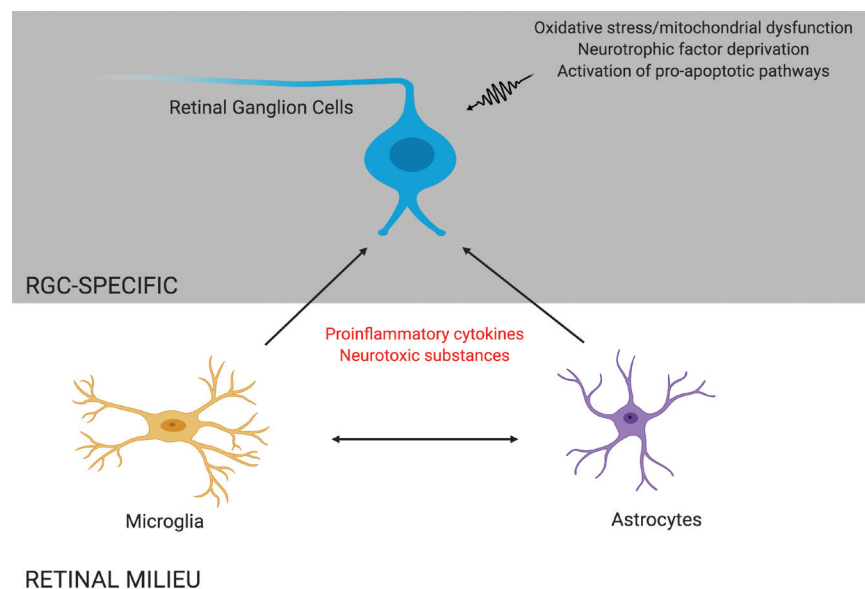


Figure 1. RGC death in glaucoma may be mediated by direct insults to RGCs, such as oxidative stress and neurotrophic factor deprivation, and by indirect proinflammatory effects caused by other retinal cells, such as microglia and astrocytes.

promise in protecting against RGC loss in animal models of glaucoma.^{8,9} A subsequent phase 3 randomized multicenter clinical trial, however, found that memantine treatment did not have a significant effect in preventing the progression of visual field loss in patients with glaucoma.¹⁰

A second example is brimonidine, a selective alpha-adrenergic receptor agonist approved for the long-term treatment of glaucoma. In a preclinical study, subcutaneous treatment with brimonidine improved RGC survival in an animal model of high IOP.¹¹ Moreover, in the Low-Pressure Glaucoma Treatment Study (LOGTS), over a 30-month period, patients treated with brimonidine had a significantly lower rate of visual field defect progression compared to patients treated with timolol (9.1% vs 39.2%), although the mean treated IOP was similar for the two groups at all time points. The results of this study and the neuroprotective efficacy of brimonidine have since been questioned because of the higher rate of patient dropout, largely due to ocular allergies, in the brimonidine group (55% vs 29% in the brimonidine and timolol groups, respectively).¹²

Other molecules that have been tested for neuroprotective qualities in preclinical studies include brain-derived neurotrophic factor, human ciliary neurotrophic factor, calcium channel blockers, antioxidants, and ginkgo biloba extract.¹³ Further clinical studies are required to determine whether they have neuroprotective effects on glaucoma patients.

WHY SO MANY CHALLENGES?

There are many reasons why it is difficult to translate preclinical data into clinical practice. First, glaucoma is a heterogeneous disease that can be difficult to fully mimic in animal models. Human clinical trials include considerable disease variability, which can be caused by multiple factors, including other medical comorbidities and polypharmacy.

Another fundamental difference between clinical trials and animal studies is the timing of intervention. In experimental studies, the neuroprotective agent is usually administered before or at the time of injury, whereas, in human clinical trials, patients are enrolled for intervention after being diagnosed with the disease.

There is also a difference in outcome measures between clinical and preclinical studies. In preclinical studies, histology is often employed to measure treatment efficacy. In contrast, in clinical trials, functional outcomes such as perimetry are frequently used, and these can take months to years to show change.

Lastly, it is often difficult to predict both the appropriate dosage of a neuroprotective agent for human use based on animal studies and the human ocular bioavailability.

NEW DRUG TARGETS

Focused on RGCs

Recent preclinical studies have focused on identifying cellular pathways that mediate RGC death to uncover new neuroprotective strategies. One study used an unbiased functional genomic screen of kinases involved in RGC death by delivering small interfering RNA to primary mouse RGCs.¹⁴ This and other work have identified dual leucine zipper kinase as playing an important role in RGC death in response to axonal injury, rendering dual leucine zipper kinase an attractive neuroprotective target.¹⁴⁻¹⁶ Subsequent high-throughput functional genomic screens have identified downstream signaling partners that may also be promising targets.¹⁷

Another study used RNA sequencing to identify molecular changes in RGCs that preceded glaucomatous neurodegeneration in a mouse model of age-related inherited glaucoma.¹⁸ The researchers showed that mitochondrial dysfunction occurred early in RGCs during glaucoma and that dietary supplementation of vitamin B3 and/or gene therapy driving expression of Nmnat1, a key enzyme producing NAD⁺, was protective against glaucoma.¹⁸

Clinical trials are needed to determine if these interventions will benefit glaucoma patients.

AT A GLANCE

- ▶ Although clinical trials have failed to show any significant benefit of candidate neuroprotective agents in glaucoma, neuroprotection holds promise as a therapeutic strategy.
- ▶ With the advent of high-throughput functional genomic screens, researchers have identified novel signaling pathways that mediate retinal ganglion cell death that can be targets for neuroprotection.
- ▶ Investigators are also studying the role of astrocytes and microglia in modulating retinal ganglion cell survival.
- ▶ Novel methodologies in trial design and imaging hold promise in translating the wealth of preclinical data into clinical practice.

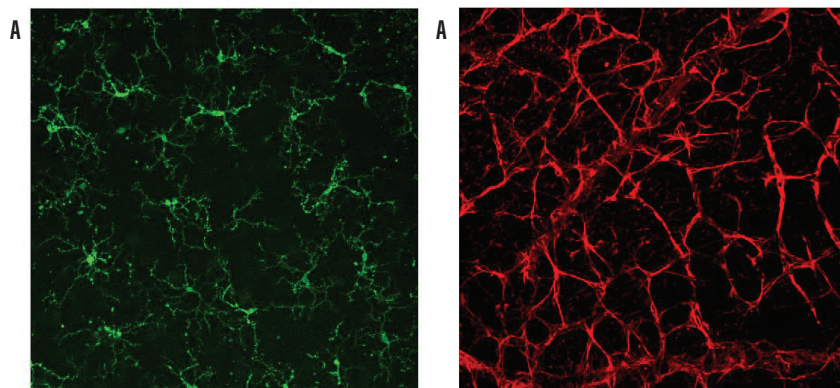


Figure 2. Mouse retina wholemounts showing retinal microglia stained with antibody against Tmem119 (panel A, green) and retinal astrocytes stained with antibody against glial fibrillary acidic protein (panel B, red).

Focused on Microglia and Astrocytes

The RGCs are contained within various cellular milieus, including the retina and the optic nerve head, which contain astrocytes, microglia and macrophages, and vasculature (Figure 2). Importantly, the survival of RGCs can depend on their interactions with these other cell types. Histopathologic studies of the glaucomatous optic nerve head have shown increased reactivity in astrocytes and microglia (resident macrophages in neural tissue), and these changes can have both protective and injurious effects on RGC survival.¹⁹ For example, in a mouse glaucoma model, ocular hypertension led to upregulation of the proinflammatory cytokine tumor necrosis factor (TNF)-alpha, followed by microglial activation and a delayed loss of RGCs.²⁰ Inhibiting TNF-alpha through the systemic administration of etanercept has been shown to inhibit microglial reactivity and prevent axonal degeneration and loss of RGCs.²¹ By secreting TNF-alpha, other cytokines and complement proinflammatory microglia can in turn stimulate astrocytes to become directly and potentially toxic to RGCs.²² These findings suggest that reducing proinflammatory cytokines and neurotoxic substances released by astrocytes and microglia may be viable neuroprotective strategies for glaucoma.

GLAUCOMA CLINICAL TRIALS: NOVEL METHODOLOGIES

Use of novel methodologies and designs in clinical trials can aid translation of preclinical data to clinical use. For example, future clinical studies can incorporate the use of OCT in measuring peripapillary retinal nerve fiber layer thickness or macular ganglion cell complex thickness as outcome measures in addition to functional outcomes such as the visual field. Novel imaging techniques of identifying RGC death in vivo via direct detection of individual apoptotic retinal cells may enable high-throughput screening of candidate neuroprotective agents and provide more sensitive outcome measures for clinical trials.^{23,24}

Although clinical trials thus far have failed to show any significant benefit of candidate neuroprotective agents in glaucoma, neuroprotection holds promise as a therapeutic strategy. ■

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