

Novel Approaches to Neuroprotection

Computer modeling seeks new agents; sustained-release technology aims to improve the efficacy of an existing drug.

BY JENNIFER KREATSOULAS, PhD, SENIOR ASSOCIATE EDITOR

Retinal neuroprotection is therapy aimed at protecting the health of photoreceptors, retinal ganglion cells, and other important neurons in the event of disease or trauma. It involves preventing or ameliorating the effects of apoptosis, programmed cell death, and necrosis, the premature death of cells and living tissue, in the retina. Research in animal models and human subjects have produced promising agents for retinal neuroprotection; however, novel neuroprotective agents and improved methods of measuring photoreceptor death and function in vivo are needed to determine how best to provide significant structural and functional rescue in the retina and prevent vision loss. Computational modeling and sustained delivery technology are two novel approaches currently undergoing evaluation to improve neuroprotection.

VIRTUAL SCREENING

Computational modeling is widely used in the pharmaceutical industry for drug discovery. Although it is infrequently applied in the retina field, Anne Hanneken, MD, and colleagues are using this technology to screen potential neuroprotective agents. In the setting of retinal detachments and retinal degenerations, the opsin molecule, which forms the visual pigment, becomes unstable and degrades. Dr. Hanneken is using computational modeling to identify alternative ligands for opsin that can potentially stabilize free opsin, prevent its degradation, and protect photoreceptors from cell death.

"These experiments are being conducted in silico, which means they are done on computer models. We validate the most promising compounds using biochemical assays that examine ligand binding in the

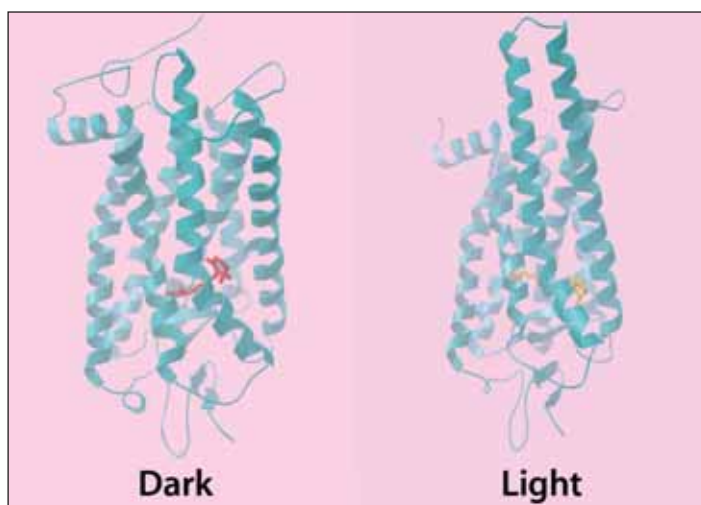


Figure 1. Computational models of the visual pigment, rhodopsin, reveal the important conformational changes that accompany the transition from the dark-adapted state to the unstable, light-activated (Meta II) state. These models are used in virtual screens to search for alternative ligands that bind to the retinal binding pocket.

opsin binding pocket and the modulation of opsin signaling," said Dr. Hanneken, an Associate Professor at the Scripps Research Institute in La Jolla, California, in an interview with *Retina Today*.

AutoDock, a computational modeling software program developed at the Scripps Research Institute, is a suite of automated docking tools designed to predict how small molecules, such as substrates or drug candidates, bind to a receptor of known 3D structure. Hundreds of computers are linked together to provide the computational power necessary to calculate these dockings. Occasionally, supercomputers are necessary to obtain the computational power that is necessary for specific molecular dynamic simulations.

"The docking program docks a particular molecule

atom by atom into the binding pocket,” Dr. Hanneken said. “All of the molecular interactions of each atom within that pocket are calculated and converted into a mathematical format that predicts the likelihood that a compound will fit in that space. The program can look at hydrogen bonds and steric interactions, which will be indicative of how likely or unlikely a compound will bind.

Opsin, a G protein-coupled receptor, activates transducin (also called Gt), which is a heterotrimeric G protein that is naturally expressed in retina rods and cones (Figure 1). Only retinoids, a well-characterized class of compounds, have been recognized as opsin-binding ligands. Retinoids typically form a covalent bond in the binding pocket of opsin.

“The idea that other compounds would dock in the opsin-binding pocket is novel. Several vision scientists thought it was highly improbable when we initially proposed this idea,” Dr. Hanneken said. “But we have shown that it does occur. Unlike retinoids, the compounds that we have discovered do not form covalent bonds, giving them a lower affinity to opsin. From our perspective, this is a distinct advantage. We don’t want to block any binding with the native chromophore. Our goal is to stabilize opsin when the chromophore is missing and prevent opsin degradation until the eye heals itself enough to restore the normal concentration of retinal in the photoreceptors.”

To date, Dr. Hanneken and colleagues have identified specific compounds with highly favorable binding free energies for the retinal binding pocket of both rod and cone opsins. Biochemical assays demonstrate that these molecules specifically dock into the retinal-binding pocket and produce opsin-mediated changes in transducin activation, similar to the interaction that certain native retinoids have with rod and cone opsins. The next step is to use these components in a variety of in vivo models and determine whether this approach will prevent photoreceptor cell death.

SUSTAINED DELIVERY

Brimonidine is a selective alpha 2-adrenergic agonist currently used for reducing intraocular pressure in ocular hypertension and glaucoma. Experimental evidence has demonstrated that brimonidine is a potential neuroprotective agent; however, clinical trials have not demonstrated efficacy in humans. According to Saylor et al,¹ an evidenced-based review of the neuroprotective qualities of brimonidine in optic nerve and retinal injury confirms that brimonidine therapy meets three of the four criteria for neuroprotection: (1) brimonidine has receptors on target tissues; (2) it adequately penetrates the vitreous and retina at pharmacologic levels, and (3) it induces intracellular changes that enhance neuronal resistance to insults or interrupt apoptosis in animal models.

Brimonidine did not meet the fourth neuroprotective criterion of success in humans.

In an effort to demonstrate neuroprotective success with brimonidine in humans, Allergan, Inc. (Irvine, CA), is currently conducting two clinical trials evaluating a new technology that administers brimonidine in a sustained delivery system similar to its intravitreal 700-μg dexamethasone implant (Ozurdex). Intravitreal brimonidine as an implant for retinal apoptosis is being explored in patients with geographic atrophy (GA) due to age-related macular degeneration (AMD) or retinitis pigmentosa (RP).

The brimonidine intravitreal implant phase 2 study in patients with GA due to AMD is ongoing. The trial² consists of two stages. Stage 1 is a patient-masked, dose-escalation, safety evaluation of brimonidine intravitreal implant. Patients will receive the implant (200 μg or 400 μg) in one eye and sham treatment in the fellow eye. Stage 2, which will begin after the safety of stage 1 has been evaluated over a 1-month period, is a randomized, double-masked, dose-response, sham-controlled evaluation of the safety and efficacy of brimonidine intravitreal implants in patients with GA due to AMD. Patients will be followed for 2 years. The trial is estimated to be completed in December 2011.

The RP study³ is an exploratory, 12-month, ascending-dose study (100 μg, 200 μg, and 400 μg) designed to evaluate the safety and effects on visual function of a single brimonidine intravitreal implant in one eye of patients with RP. The trial is estimated to be completed in July 2010.

CONCLUSION

Allergan’s clinical trials, along with ongoing research by Dr. Hanneken, and Cagri Besirli, MD, PhD, and David N. Zacks, MD, PhD, who contributed to the March 2010 issue of *Retina Today* with an article titled “Retinal Neuroprotection,” offer innovative approaches to identifying retinal neuroprotective agents. As Drs. Besirli and Zacks suggested, successful neuroprotection may require combination treatments of anti-apoptotics and exogenous pro-survival agents. Innovative technologies, such as computer modeling and sustained delivery systems, hold much promise for contributing to successful neuroprotection in the setting of retinal diseases. ■

1. Saylor M, McLoon LK, Harrison, AR, Lee MS. Experimental and clinical evidence for brimonidine as an optic nerve and retinal neuroprotective agent: an evidence-based review. *Arch Ophthalmol*. 2009;127(4):402-406.

2. Clinicaltrials.gov. Safety and efficacy of brimonidine intravitreal implant in patients with geographic atrophy due to age-related macular degeneration (AMD). Available at: <http://www.clinicaltrial.gov/ct2/show/NCT00658619?term=brimonidine&rank=3>. Accessed March 25, 2010.

3. Clinicaltrials.gov. An exploratory study to evaluate the safety of brimonidine intravitreal implant in patients with retinitis pigmentosa. Available at: <http://www.clinicaltrial.gov/ct2/show/NCT00661479?term=brimonidine&rank=13>. Accessed March 25, 2010.