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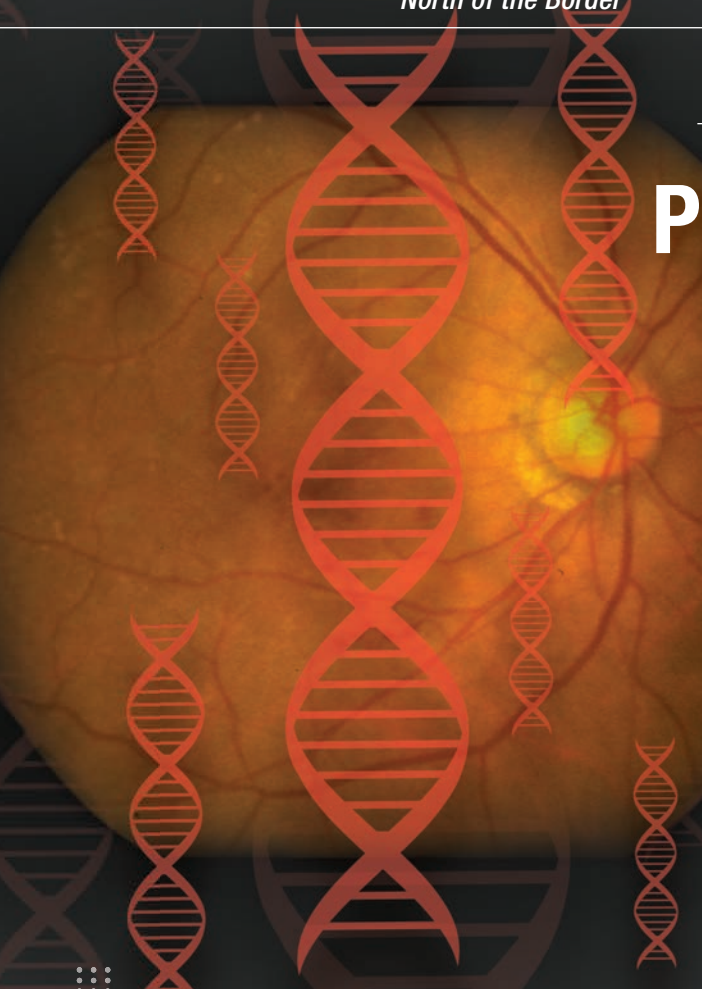
RETINA SPECIALIST

MARCH 2017



Gene Therapy and Cutting-Edge Science

PHARMACOGENETICS and AMD

What we know (and don't know) so far.

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For Inherited Retinal Disease

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Gene Therapy: New Frontier
For Inherited Retinal Disease

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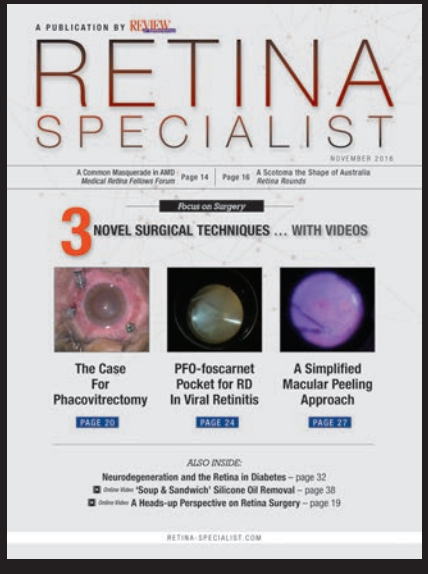

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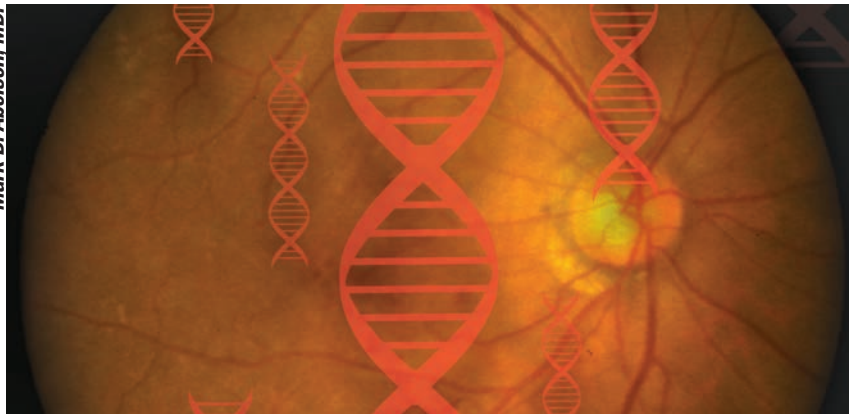
MARCH 2017

FEATURES



Mark B. Abelson, MD.

Gene Therapy and Cutting-Edge Science

DEPARTMENTS

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LUCENTIS®
RANIBIZUMAB INJECTION

INDICATIONS

LUCENTIS is indicated for the treatment of patients with:

- Neovascular (wet) age-related macular degeneration (wAMD)
- Macular edema following retinal vein occlusion (RVO)

IMPORTANT SAFETY INFORMATION

CONTRAINDICATIONS

- LUCENTIS is contraindicated in patients with ocular or periocular infections or hypersensitivity to ranibizumab or any of the excipients in LUCENTIS

WARNINGS AND PRECAUTIONS

- Intravitreal injections, including those with LUCENTIS, have been associated with endophthalmitis, retinal detachment, and iatrogenic traumatic cataract. Proper aseptic injection technique should always be utilized when administering LUCENTIS. Patients should be monitored following the injection to permit early treatment, should an infection occur
- Increases in intraocular pressure (IOP) have been noted both pre-injection and post-injection (at 60 minutes) with LUCENTIS. IOP and perfusion of the optic nerve head should be monitored and managed appropriately
- Although there was a low rate of arterial thromboembolic events (ATEs) observed in the LUCENTIS clinical trials, there is a potential risk of ATEs following intravitreal use of VEGF inhibitors. ATEs are defined as nonfatal stroke, nonfatal myocardial infarction, or vascular death (including deaths of unknown cause)

ADVERSE EVENTS

- Serious adverse events related to the injection procedure that occurred in <0.1% of intravitreal injections included endophthalmitis, rhegmatogenous retinal detachment, and iatrogenic traumatic cataract
- In the LUCENTIS Phase III clinical trials, the most common ocular side effects included conjunctival hemorrhage, eye pain, vitreous floaters, increased intraocular pressure, vitreous detachment, and intraocular inflammation and maculopathy. The most common non-ocular side effects included nasopharyngitis, headache, arthralgia, bronchitis, influenza, and sinusitis

Please see Brief Summary of LUCENTIS full Prescribing Information on adjacent page.

*The following randomized, double-masked pivotal trials were conducted for the 2 LUCENTIS indications: **wAMD:** *MARINA*—Phase III, multicenter, 2-year, sham injection–controlled study; primary end point at 1 year. *ANCHOR*—Phase III, multicenter, 2-year, active treatment–controlled study; primary end point at 1 year. *PIER*—Phase IIIb, 2-year, sham injection–controlled study; primary end point at 1 year. *HARBOR*—Phase III, multicenter, 2-year, active treatment–controlled dose-response study; primary end point at 1 year. **RVO:** *BRAVO*—Phase III, multicenter, 1-year, sham injection–controlled study; primary end point at 6 months. *CRUISE*—Phase III, multicenter, 1-year, sham injection–controlled study; primary end point at 6 months.[1-7]

REFERENCES: 1. Rosenfeld PJ, et al; MARINA Study Group. *N Engl J Med.* 2006;355:1419-1431. **2.** Brown DM, et al; ANCHOR Study Group. *Ophthalmology.* 2009;116:57-65. **3.** Regillo CD, et al; PIER Study Group. *Am J Ophthalmol.* 2008;145:239-248. **4.** Busbee BG, et al; HARBOR Study Group. *Ophthalmology.* 2013;120:1046-1056. **5.** Campochiaro PA, et al; BRAVO Investigators. *Ophthalmology.* 2010;117:1102-1112. **6.** Brown DM, et al; CRUISE Investigators. *Ophthalmology.* 2010;117:1124-1133. **7.** LUCENTIS [package insert]. South San Francisco, CA: Genentech, Inc; 2016.

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LUCENTIS®
RANIBIZUMAB INJECTION

Brief summary–please see the LUCENTIS® package insert for full prescribing information.

1 INDICATIONS AND USAGE
LUCENTIS is indicated for the treatment of patients with:

1.1 Neovascular (wet) age-related macular degeneration (AMD)

1.2 Macular edema following retinal vein occlusion (RVO)

1.3 Diabetic macular edema (DME)

1.4 Diabetic retinopathy (non proliferative diabetic retinopathy (NPDR), proliferative diabetic retinopathy (PDR)) in patients with diabetic macular edema (DME)

4 CONTRAINDICATIONS
4.1 Ocular or periocular infections
LUCENTIS is contraindicated in patients with ocular or periocular infections.

4.2 Hypersensitivity
LUCENTIS is contraindicated in patients with known hypersensitivity to ranibizumab or any of the excipients in LUCENTIS. Hypersensitivity reactions may manifest as severe intraocular inflammation.

5 WARNINGS AND PRECAUTIONS
5.1 Endophthalmitis and retinal detachments
Intravitreal injections, including those with LUCENTIS, have been associated with endophthalmitis and retinal detachments. Proper aseptic injection technique should always be used when administering LUCENTIS. In addition, patients should be monitored following the injection to permit early treatment should an infection occur *[see Patient Counseling Information (17)]*.

5.2 Increases in intraocular pressure
Increases in intraocular pressure have been noted both pre-injection and post-injection (at 60 minutes) while being treated with LUCENTIS. Monitor intraocular pressure prior to and following intravitreal injection with LUCENTIS and manage appropriately.

5.3 Thromboembolic events
Although there was a low rate of arterial thromboembolic events (ATEs) observed in the LUCENTIS clinical trials, there is a potential risk of ATEs following intravitreal use of VEGF inhibitors. ATEs are defined as nonfatal stroke, nonfatal myocardial infarction, or vascular death (including deaths of unknown cause).

Neovascular (wet) age-related macular degeneration
The ATE rate in the three controlled neovascular AMD studies (AMD-1, AMD-2, AMD-3) during the first year was 1.9% (17 of 874) in the combined group of patients treated with 0.3 mg or 0.5 mg LUCENTIS compared with 1.1% (5 of 441) in patients from the control arms *[see Clinical Studies (14.1)]*. In the second year of Studies AMD-1 and AMD-2, the ATE rate was 2.6% (19 of 721) in the combined group of LUCENTIS-treated patients compared with 2.9% (10 of 344) in patients from the control arms. In Study AMD-4, the ATE rates observed in the 0.5 mg arms during the first and second year were similar to rates observed in Studies AMD-1, AMD-2, and AMD-3.

In a pooled analysis of 2-year controlled studies (AMD-1, AMD-2, and a study of LUCENTIS used adjunctively with verteporfin photodynamic therapy), the stroke rate (including both ischemic and hemorrhagic stroke) was 2.7% (13 of 484) in patients treated with 0.5 mg LUCENTIS compared to 1.1% (5 of 435) in patients in the control arms (odds ratio 2.2 (95% confidence interval (0.8-7.1))).

Macular edema following retinal vein occlusion
The ATE rate in the two controlled RVO studies during the first 6 months was 0.8% in both the LUCENTIS and control arms of the studies (4 of 525 in the combined group of patients treated with 0.3 mg or 0.5 mg LUCENTIS and 2 of 260 in the control arms) *[see Clinical Studies (14.2)]*. The stroke rate was 0.2% (1 of 525) in the combined group of LUCENTIS-treated patients compared to 0.4% (1 of 260) in the control arms.

Diabetic macular edema and diabetic retinopathy
Safety data are derived from studies D-1 and D-2. All enrolled patients had DME and DR at baseline *[see Clinical Studies (14.3, 14.4)]*.

In a pooled analysis of Studies D-1 and D-2 *[see Clinical Studies (14.3)]*, the ATE rate at 2 years was 7.2% (18 of 250) with 0.5 mg LUCENTIS, 5.6% (14 of 250) with 0.3 mg LUCENTIS, and 5.2% (13 of 250) with control. The stroke rate at 2 years was 3.2% (8 of 250) with 0.5 mg LUCENTIS, 1.2% (3 of 250) with 0.3 mg LUCENTIS, and 1.6% (4 of 250) with control. At 3 years, the ATE rate was 10.4% (26 of 249) with 0.5 mg LUCENTIS and 10.8% (27 of 250) with 0.3 mg LUCENTIS; the stroke rate was 4.8% (12 of 249) with 0.5 mg LUCENTIS and 2.0% (5 of 250) with 0.3 mg LUCENTIS.

5.4 Fatal events in patients with DME and DR at baseline
Diabetic macular edema and diabetic retinopathy
Safety data are derived from studies D-1 and D-2. All enrolled patients had DME and DR at baseline *[see Clinical Studies (14.3, 14.4)]*.

A pooled analysis of Studies D-1 and D-2 *[see Clinical Studies (14.3)]*, showed that fatalities in the first 2 years occurred in 4.4% (11 of 250) of patients treated with 0.5 mg LUCENTIS, in 2.8% (7 of 250) of patients treated with 0.3 mg LUCENTIS, and in 1.2% (3 of 250) of control patients. Over 3 years, fatalities occurred in 6.4% (16 of 249) of patients treated with 0.5 mg LUCENTIS and in 4.4% (11 of 250) of patients treated with 0.3 mg LUCENTIS. Although the rate of fatal events was low and included causes of death typical of patients with advanced diabetic complications, a potential relationship between these events and intravitreal use of VEGF inhibitors cannot be excluded.

6 ADVERSE REACTIONS
The following adverse reactions are discussed in greater detail in the *Warnings and Precautions (5)* section of the label:
- Endophthalmitis and retinal detachments
- Increases in intraocular pressure
- Thromboembolic events
- Fatal events in patients with DME and DR at baseline

6.1 Injection procedure
Serious adverse reactions related to the injection procedure have occurred in < 0.1% of intravitreal injections, including endophthalmitis *[see Warnings and Precautions (5.1)]*, rhegmatogenous retinal detachment, and iatrogenic traumatic cataract.

6.2 Clinical studies experience
Because clinical trials are conducted under widely varying conditions, adverse reaction rates observed in one clinical trial of a drug cannot be directly compared with rates in the clinical trials of the same or another drug and may not reflect the rates observed in practice.

The data below reflect exposure to 0.5 mg LUCENTIS in 440 patients with neovascular AMD in Studies AMD-1, AMD-2, and AMD-3, and 259 patients with macular edema following RVO. The data also reflect exposure to 0.3 mg LUCENTIS in 250 patients with DME and DR at baseline *[see Clinical Studies (14)]*.

Safety data observed in Study AMD-4 were consistent with these results. On average, the rates and types of adverse reactions in patients were not significantly affected by dosing regimen.

Ocular reactions
Table 1 shows frequently reported ocular adverse reactions in LUCENTIS-treated patients compared with the control group.

Table 1 Ocular reactions in the DME and DR, AMD, and RVO studies

	DME and DR 2-year		AMD 2-year		AMD 1-year		RVO 6-month	
	LUCENTIS 0.3 mg	Control	LUCENTIS 0.5 mg	Control	LUCENTIS 0.5 mg	Control	LUCENTIS 0.5 mg	Control
Adverse Reaction	n=250	n=250	n=379	n=379	n=440	n=441	n=259	n=260
Conjunctival hemorrhage	47%	32%	74%	60%	64%	50%	48%	37%
Eye pain	17%	13%	35%	30%	26%	20%	17%	12%
Vitreous floaters	10%	4%	27%	8%	19%	5%	7%	2%
Intraocular pressure increased	18%	7%	24%	7%	17%	5%	7%	2%
Vitreous detachment	11%	15%	21%	19%	15%	15%	4%	2%
Intraocular inflammation	4%	3%	18%	8%	13%	7%	1%	3%
Cataract	28%	32%	17%	14%	11%	9%	2%	2%
Foreign body sensation in eyes	10%	5%	16%	14%	13%	10%	7%	5%
Eye irritation	8%	5%	15%	15%	13%	12%	7%	6%
Lacrimation increased	5%	4%	14%	12%	8%	8%	2%	3%
Blepharitis	3%	2%	12%	8%	8%	5%	0%	1%
Dry eye	5%	3%	12%	7%	7%	7%	3%	3%
Visual disturbance or vision blurred	8%	4%	18%	15%	13%	10%	5%	3%
Eye pruritis	4%	4%	12%	11%	9%	7%	1%	2%
Ocular hyperemia	9%	9%	11%	8%	7%	4%	5%	3%
Retinal disorder	2%	2%	10%	7%	8%	4%	2%	1%
Maculopathy	5%	7%	9%	9%	6%	6%	11%	7%
Retinal degeneration	1%	0%	8%	6%	5%	3%	1%	0%
Ocular discomfort	2%	1%	7%	4%	5%	2%	2%	2%
Conjunctival hyperemia	1%	2%	7%	6%	5%	4%	0%	0%
Posterior capsule opacification	4%	3%	7%	4%	2%	2%	0%	1%
Injection site hemorrhage	1%	0%	5%	2%	3%	1%	0%	0%

Non-ocular reactions

Non-ocular adverse reactions with an incidence of ≥ 5% in patients receiving LUCENTIS for DR, DME, AMD, and/or RVO and which occurred at a ≥ 1% higher frequency in patients treated with LUCENTIS compared to control are shown in Table 2. Though less common, wound healing complications were also observed in some studies.

Table 2 Non-ocular reactions in the DME and DR, AMD and RVO studies

	DME and DR 2-year		AMD 2-year		AMD 1-year		RVO 6-month	
	LUCENTIS 0.3 mg	Control	LUCENTIS 0.5 mg	Control	LUCENTIS 0.5 mg	Control	LUCENTIS 0.5 mg	Control
Adverse Reaction	n=250	n=250	n=379	n=379	n=440	n=441	n=259	n=260
Nasopharyngitis	12%	6%	16%	13%	8%	9%	5%	4%
Anemia	11%	10%	8%	7%	4%	3%	1%	1%
Nausea	10%	9%	9%	6%	5%	5%	1%	2%
Cough	9%	4%	9%	8%	5%	4%	1%	2%
Constipation	8%	4%	5%	7%	3%	4%	0%	1%
Seasonal allergy	8%	4%	4%	4%	2%	2%	0%	2%
Hypercholesterolemia	7%	5%	5%	5%	3%	2%	1%	1%
Influenza	7%	3%	7%	5%	3%	2%	3%	2%
Renal failure	7%	6%	1%	1%	0%	0%	0%	0%
Upper respiratory tract infection	7%	7%	9%	8%	5%	5%	2%	2%
Gastroesophageal reflux disease	6%	4%	4%	6%	3%	4%	1%	0%
Headache	6%	8%	12%	9%	6%	5%	3%	3%
Edema peripheral	6%	4%	3%	5%	2%	3%	0%	1%
Renal failure chronic	6%	2%	0%	1%	0%	0%	0%	0%
Neuropathy peripheral	5%	3%	1%	1%	1%	0%	0%	0%
Sinusitis	5%	8%	8%	7%	5%	5%	3%	2%
Bronchitis	4%	4%	11%	9%	6%	5%	0%	2%
Atrial fibrillation	3%	3%	5%	4%	2%	2%	1%	0%
Arthralgia	3%	3%	11%	9%	5%	5%	2%	1%
Chronic obstructive pulmonary disease	1%	1%	6%	3%	3%	1%	0%	0%
Wound healing complications	1%	0%	1%	1%	1%	0%	0%	0%

6.3 Immunogenicity
As with all therapeutic proteins, there is the potential for an immune response in patients treated with LUCENTIS. The immunogenicity data reflect the percentage of patients whose test results were considered positive for antibodies to LUCENTIS in immunoassays and are highly dependent on the sensitivity and specificity of the assays.

The pre-treatment incidence of immunoreactivity to LUCENTIS was 0%-5% across treatment groups. After monthly dosing with LUCENTIS for 6 to 24 months, antibodies to LUCENTIS were detected in approximately 1%-9% of patients.

The clinical significance of immunoreactivity to LUCENTIS is unclear at this time. Among neovascular AMD patients with the highest levels of immunoreactivity, some were noted to have iritis or vitritis. Intraocular inflammation was not observed in patients with DME and DR at baseline, or RVO patients with the highest levels of immunoreactivity.

6.4 Postmarketing Experience
The following adverse reactions have been identified during post-approval use of LUCENTIS. Because these reactions are reported voluntarily from a population of uncertain size, it is not always possible to reliably estimate their frequency or establish a causal relationship to drug exposure.

- Ocular: Tear of retinal pigment epithelium among patients with neovascular AMD

7 DRUG INTERACTIONS
Drug interaction studies have not been conducted with LUCENTIS.

LUCENTIS intravitreal injection has been used adjunctively with verteporfin photodynamic therapy (PDT). Twelve (12) of 105 (11%) patients with neovascular AMD developed serious intraocular inflammation; in 10 of the 12 patients, this occurred when LUCENTIS was administered 7 days (± 2 days) after verteporfin PDT.

8 USE IN SPECIFIC POPULATIONS
8.1 Pregnancy
There are no studies of LUCENTIS in pregnant women. An embryo-fetal developmental toxicity study was performed on pregnant cynomolgus monkeys. Pregnant animals received intravitreal injections of ranibizumab every 14 days starting on Day 20 of gestation, until Day 62 at doses of 0, 0.125, and 1 mg/eye. Skeletal abnormalities including incomplete and/or irregular ossification of bones in the skull, vertebral column, and hindlimbs and shortened supernumerary ribs were seen at a low incidence in fetuses from animals treated with 1 mg/eye of ranibizumab. The 1 mg/eye dose resulted in trough serum ranibizumab levels up to 13 times higher than predicted C_{max} levels with single eye treatment in humans. No skeletal abnormalities were seen at the lower dose of 0.125 mg/eye, a dose which resulted in trough exposures equivalent to single eye treatment in humans. No effect on the weight or structure of the placenta, maternal toxicity, or embryotoxicity was observed.

Animal reproduction studies are not always predictive of human response. It is also not known whether ranibizumab can cause fetal harm when administered to a pregnant woman or can affect reproduction capacity. Based on the anti-VEGF mechanism of action for ranibizumab *[see Clinical Pharmacology (12.1)]*, treatment with LUCENTIS may pose a risk to embryo-fetal development (including teratogenicity) and reproductive capacity. LUCENTIS should be given to a pregnant woman only if clearly needed.

8.3 Nursing mothers
It is not known whether ranibizumab is excreted in human milk. Because many drugs are excreted in human milk, and because the potential for absorption and harm to infant growth and development exists, caution should be exercised when LUCENTIS is administered to a nursing woman.

8.4 Pediatric use
The safety and effectiveness of LUCENTIS in pediatric patients have not been established.

8.5 Geriatric use
In the clinical studies, approximately 79% (2387 of 3005) of patients randomized to treatment with LUCENTIS were ≥ 65 years of age and approximately 54% (1636 of 3005) were ≥ 75 years of age *[see Clinical Studies (14)]*. No notable differences in efficacy or safety were seen with increasing age in these studies. Age did not have a significant effect on systemic exposure.

10 OVERDOSAGE
More concentrated doses as high as 2 mg ranibizumab in 0.05 mL have been administered to patients. No additional unexpected adverse reactions were seen.

17 PATIENT COUNSELING INFORMATION
Advise patients that in the days following LUCENTIS administration, patients are at risk of developing endophthalmitis. If the eye becomes red, sensitive to light, painful, or develops a change in vision, advise the patient to seek immediate care from an ophthalmologist *[see Warnings and Precautions (5.1)]*.

LUCENTIS® [ranibizumab injection]
Manufactured by:
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94080-4990

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By Charles C. Wykoff, MD, PhD

'Genetics, The Future is Now'

So *Time* magazine stated boldly on its January 1994 cover, claiming, "New breakthroughs can cure disease and save lives."

So, how have we done? Where are we from a retinal perspective?

Without a doubt, we have covered a lot of ground. Over the last two decades, more than 200 distinct genes have been identified that, when mutated, can lead to retinal dystrophies. Spring boarding from this work, multiple ongoing trials are investigating various gene-based therapies to treat a range of monogenetic retinal diseases. The approach most likely to reach clinical application first, gene-based therapy for RPE65-associated Leber congenital amaurosis, also known as LCA2, has been submitted for regulatory review in the United States with approval anticipated for this year. See page 35, where Christine Kay, MD, explores this exciting space and compares ongoing investigations in inherited retinal disease.

We have also seen great progress on retinal diseases with more complex genetic underpinnings. We have learned that the risk of age-related macular degeneration is largely attributable to our genes, although it is a complex relationship involving more than 34 genetic loci. See page 26, where Steve Schwartz, MD, MBA, and Parth Shah bring us up to date on the pharmacogenetics of AMD and how this might impact our management recommendations.

But clearly, much more remains to be learned. The American Academy of Ophthalmology continues to recommend against routine genetic testing for complex genetic diseases including AMD. Why, and when might the AAO recommend otherwise? See page 31, where Karmen Trzupek, MS, a certified genetic counselor, beautifully explores the realities and challenges of genetic testing in your clinic.

As well highlighted in an excellent 2015 *American Journal of Ophthalmology* "Perspective," gene therapy has been hailed as "five years away" from clinical application since the 1990s.[1] As clinicians, it is our responsibility to educate patients about their options while providing hope of treatments yet to come.

However, this hope must be grounded in an understanding of the reality of current research. Fortunately, the promise of clinically ready and clinically valuable genetic testing, as well as gene-based therapies, appears to be closer than ever to reality. I hope the articles to follow bring you up to date on our current understanding of the genetics underlying many of the retinal diseases we manage.

Reference

1. Benjaminy S, Kowal SP, MacDonald IM, Bubela T. Communicating the promise for ocular gene therapies: Challenges and recommendations. Am J Ophthalmol. 2015;160:408-415.

Surgical Robot Guides Microneedle in RVO

Months after surgeons in the United Kingdom completed the first robotic retina surgery to perform macular peel and place a gene vector, researchers in Belgium have used a surgical robot to place a microneedle into a retinal vein to inject a thrombolytic agent to treat retinal vein occlusion.

This surgical robot guided a 0.03-mm needle into the occluded retinal vein.

Researchers at the University Hospitals Leuven and the Catholic University Leuven (known as KU Leuven for its Dutch spelling) developed the robot specifically for this procedure. The surgeons used the robot to guide placement of a 0.03-mm needle into the vein.

According to the research team, led by Peter Stalmans, MD, at University Hospital Leuven, and Dominiek Reynaerts, PhD, chair of mechanical engineering at KU Leuven, the procedure successfully treated the occlusion. The current Phase I trial aims to demonstrate the technical feasibility of using a robotic device to insert a microneedle into the retinal vein to inject ocriplasmin.

The procedure could be a cost-effective alternative to existing treatments, Dr. Stalmans says. "Current treatment for retinal vein occlusion costs 32,000 euro per eye (about $34,000), a high price tag, especially if you know that you are only treating the side effects and that there is little more you can do than avoid decreasing eye sight," he says. "The robotic device enables us to treat the cause of the thrombosis in the retina for the first time."

Dr. Reynaerts says the goal is to use the robotic device to perform other procedures.

The next step is to conduct a Phase II trial to determine if the procedure is clinically effective for RVO.

Late last year, surgeons at John Radcliffe Hospital in Oxford, U.K., performed the first robotic retina operation using a device developed by the Dutch medical robotics firm Preceyes BV.

IN BRIEF

• **GenSight Biologics** received orphan drug designation from the Food and Drug Administration for its candidate **GS030** for treatment of retinitis pigmentosa. GenSight expects to start a Phase I/II clinical trial in RP in the third quarter.

• The FDA granted **Genentech** approval for **Lucentis** (ranibizumab) for treatment of myopic choroidal neovascularization. This is the fifth approved indication for Lucentis since its launch 2006.

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A Twist on Ocular Trauma

Long bone fracture leads to a curious retinopathy.

By Adam Sweeney, MD

An 18-year-old white male college student with history of Duchenne muscular dystrophy presented to the Seattle Children's Hospital emergency room after suffering an accidental ground-level fall with subsequent vision loss.

The patient was hospitalized with a fractured femur and fat emboli to the lungs bilaterally. He'd reported normal vision until the day after the fall, when he developed photopsias for several hours and subsequent bilateral central scotomas with reduced visual acuity.

The patient's ocular history included bilateral cataracts with baseline vision of 20/50 OD and 20/30 OS, attributed to prior use of systemic corticosteroids. He had never had ocular surgery or taken any ocular medications. Aside from restricted mobility related to his Duchenne muscular dystrophy (DMD), his medical history was otherwise unremarkable.

Examination

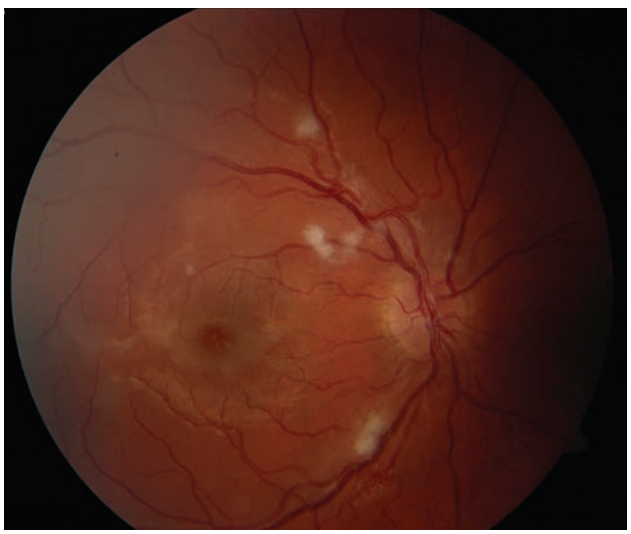
Best-corrected visual acuity was 20/60 and 20/40 with intraocular pressures of 16 mm Hg and 24 mm Hg in the right and left eyes, respectively. No afferent pupillary defect was present, visual fields were full to confrontation and ocular motility was normal in both eyes. The anterior segment was normal with exception of bilateral 3+ central posterior subcapsular cataracts and cortical lenticular opacities.

The vitreous was normal bilaterally. Fundus examination of the right eye revealed cotton wool spots (CWS) located primarily along the superior and inferior arcades adjacent to but not overlying the retinal arterioles. A few small CWS along the vascular arcades were evident nasally. Additionally, the fovea had a grayish opaque appearance with loss of the normal foveal light reflex. The dilated fundus exam of the left eye showed a similar appearance (*Figure 1*). No hemorrhages or optic nerve head edema were noted.

Workup, Diagnosis And Management

Because the patient could not sit for a desktop optical coherence tomography (OCT) test due to his injuries, we performed a handheld OCT that demonstrated no gross intraretinal or subretinal fluid but did show hyper-reflective intraretinal lesions (*Figure 2, page 10*). We deferred further workup given the strong clinical suspicion for fat embolism to the retinal microvasculature leading to a Purtscher-like retinopathy.

After his hospital discharge, we evaluated the patient at the Uni-



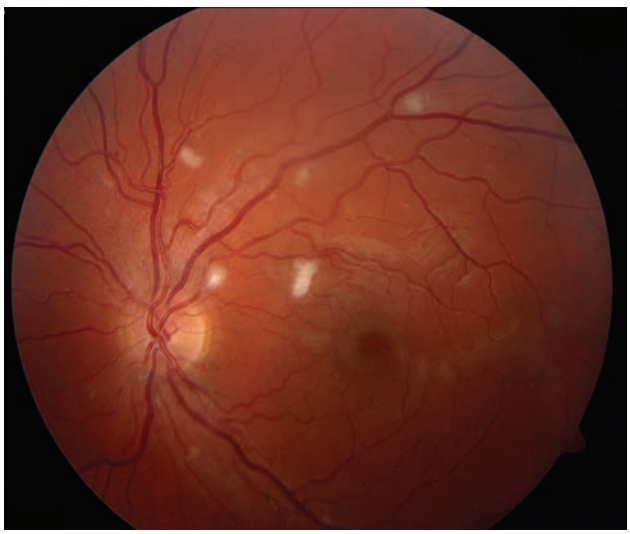


Figure 1. Color fundus photographs show bilateral cotton wool spots near the vascular arcades in both eyes as well as loss of the foveal light reflex and grayish opacification of the retina near the fovea.

versity of Washington Eye Institute clinic, where we documented fewer and smaller CWS since discharge as well as improved visual acuity. We did not obtain fluorescein angiography due to patient positioning restrictions and strong clinical suspicion. We arranged for monitoring with clinical exams at one, two and six months post-injury per the published recommendations for patients with this condition.[1]

Discussion

Purtscher-like retinopathy is a rare occlusive vasculopathy. The classical manifestations are bilateral white retinal lesions, CWS and posterior-pole hemorrhage.

Differentiated from Purtscher's retinopathy, originally described as following compressive trauma to the head or chest, Purtscher-like retinopathy can arise from other systemic insults. These include acute pancreatitis, fat embolism syndrome, chronic renal failure, amniotic fluid embolism from childbirth, connective tissue disorders, cryoglobulinemia, weightlifting, shaken-baby syndrome, retrobulbar anesthesia and orbital steroid injection, among other causes.[2]

Clinically, patients present with decreased visual acuity noted at 24 to 48 hours after the injury. Visual acuity is typically better than hand motions in both eyes with central or paracentral visual field loss.

The etiology of Purtscher's and Purtscher-like retinopathy is not well understood, but is likely secondary to embolic occlusion of arterioles. However, authors have proposed numerous etiologies, including vasculitis, raised intrathoracic pressure or, as Otmar Purtscher originally postulated, raised intracranial pressure and extravasation of lymphatic material from the retinal vessels.[3]

The physical examination of these patients typically features bilateral, multiple peripapillary CWS and superficial hemorrhages adjacent to the retinal arterioles. Purtscher flecken are polygonal shaped superficial retinal white lesions with a clear zone adjacent to vessels and are considered pathognomonic, but only occur in approximately 50 percent of cases.[1, 4]

Less common findings include optic disc swelling, serous macular detachment, dilated tortuous vessels, hard exudates, optic disc edema and relative afferent pupillary defects.

Findings typically lag behind the inciting event by 24 to 48 hours, similar to the onset of symptoms. The CWS of Purtscher-like retinopathy due to fat embolism, as in this case, are generally smaller and located more peripherally in the retina than in classic Purtscher's retinopathy.[5]

Imaging and Systemic Workup

Fluorescein angiography demonstrates nonperfusion of the small retinal capillaries in the regions corresponding to the retinal whitening, and, in some patients, delayed filling of vessels, late leakage and peripapillary leakage.[1] OCT may demonstrate hyper-reflectivity at the inner plexiform layer and inner nuclear layer, as it did in our case.[6]

In cases without trauma but with a characteristic fundus appearance, the systemic workup should include a detailed history and physical exam in addition to a basic metabolic panel, amylase, lipase, complete blood count and imaging as indicated.

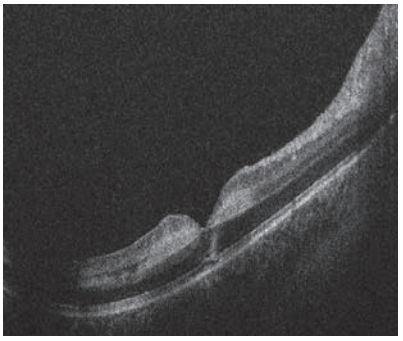


Figure 2. Handheld optical coherence tomography imaging of the macula (right eye shown here) demonstrated foveal contour preservation with alteration of the normal retinal architecture. Imaging showed a hyper-reflective lesion of the inner nuclear and outer plexiform layers, but no gross intraretinal or subretinal fluid.

Purtscher's and Purtscher-like retinopathy are clinical diagnoses, thus, a working differential is in order.

Other conditions with a similar fundus appearance include central retinal vein occlusion, central retinal artery occlusion and *commotio retinae*. Supportive care with treatment of the underlying condition is the only recommended management. While some authors have reported treatment with systemic steroids, controlled studies are lacking and retrospective reports have not shown statistically significant improvement in vision.[1, 3]

Retinal lesions generally resolve spontaneously within one to three months, replaced with attenuation of vessels, temporal disc pallor or mottling of the retinal pigment epithelium.[2] Visual acuity recovery is variable, but returns to baseline in at least 50 percent of patients, with a better prognosis for patients without associated macular edema.[1]

(Continued on page 13)

By Fang Zheng, MD, and
Philip J. Rosenfeld, MD, PhD

SD- vs. SS-OCTA for CNV in NVAMD

Here's a look at how the two techniques compare for detecting macular neovascularization.

Optical coherence tomography has become an invaluable imaging technique for diagnosing and following patients with neovascular age-related macular degeneration.[1] However, up until recently, OCT imaging could only indirectly detect the macular neovascularization by showing structural changes in the macula and the excess production of vascular endothelial growth factor.

These changes include increased reflectivity from the outer retina to Bruch's membrane and the accumulation of fluid in and under the retina, and under the retinal pigment epithelium (RPE).

To directly visualize the macular neovascularization (MNV), we needed to perform invasive dye-based angiographic techniques such as fluorescein angiography and indocyanine angiography. Now, with the development of OCT angiography (OCTA), it is possible to directly image the MNV in the retina (type 3), under the retina and above the RPE (type 2), and under the RPE (type 1).

What is OCTA?

OCTA detects blood flow by performing multiple B-scans at the same position, and the algorithms generate a decorrelation signal based on changes in the intensity and/or phase information between repeated OCT B-scans. This decorrelation signal, which reflects subtle differences between B-scans at the same position, results mostly from the movement of erythrocytes within blood vessels.

While different OCTA instruments use different hardware configurations and different decorrelation algorithms, the basic premise for the detection of blood flow remains the same for all the different OCTA imaging strategies.

Nowadays, two main types of OCTA instruments are used to detect MNV: spectral-domain OCTA (SD-OCTA) and swept-source OCTA (SS-OCTA). Both use Fourier domain detection techniques, but the SD-OCT instruments use a broadband near-infrared super luminescent diode as a light source, currently with a center wavelength of approximately 840 nm, with a spectrometer as the detector, while SS-OCT devices use a tunable swept laser, currently with a center wavelength of approximately 1,050 nm, with a single photodiode detector.[2]

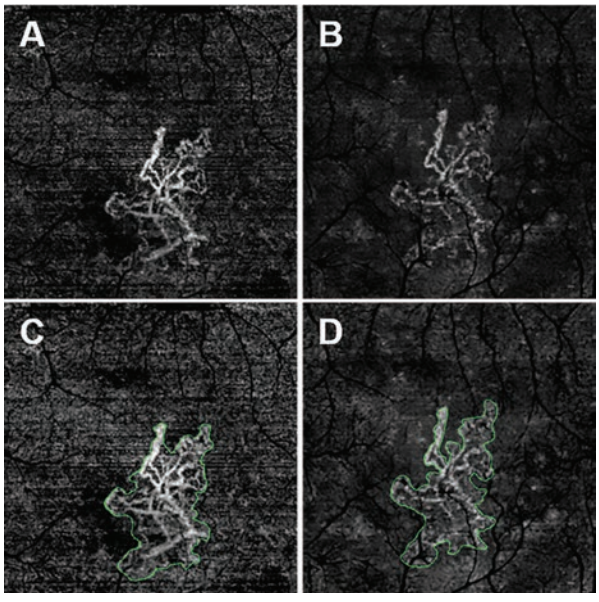


Figure 1. En face swept-source (SS) and spectral-domain (SD) optical coherence tomography angiography (OCTA) images of the left eye from an 81-year-old man with macular neovascularization (MNV) secondary to age-related macular degeneration. All images were processed from the corresponding volumetric datasets using the same algorithms applied to a slab from the outer retina to the choriocapillaris with removal of the projection artifacts from the retinal vasculature. SS-OCTA 3-mm × 3-mm scan (A); SD-OCTA 3-mm × 3-mm scan (B); SS-OCTA 3-mm × 3-mm scan with an outline of the MNV and an area of 1.029 mm2 (C); and SD-OCTA 3-mm × 3-mm scan with an outline of the choroidal neovascularization and an area of 0.958 mm² (D).

Which Is Better For Detecting MNV?

The main advantage of SS-OCTA imaging over SD-OCTA is a faster scanning speed, which allows for denser scan patterns and larger scan areas than SD-OCTA scans for a given acquisition time.

Another advantage of the current SS-OCTA technology is that it uses a longer center wavelength that can reduce sensitivity roll-off of the signal under the RPE, which results in enhanced light penetration into the choroid and better detection of signals from the deeper layers.

Additionally, the longer wavelength of SS-OCTA is safer for the eye and allows for the use of a higher laser power.

All of this improves the likelihood that SS-OCTA will detect the inherently weaker signals from deeper layers of the retina. Overall, these advantages should help the SS-OCT system overcome the barrier of the RPE, resulting in better de-

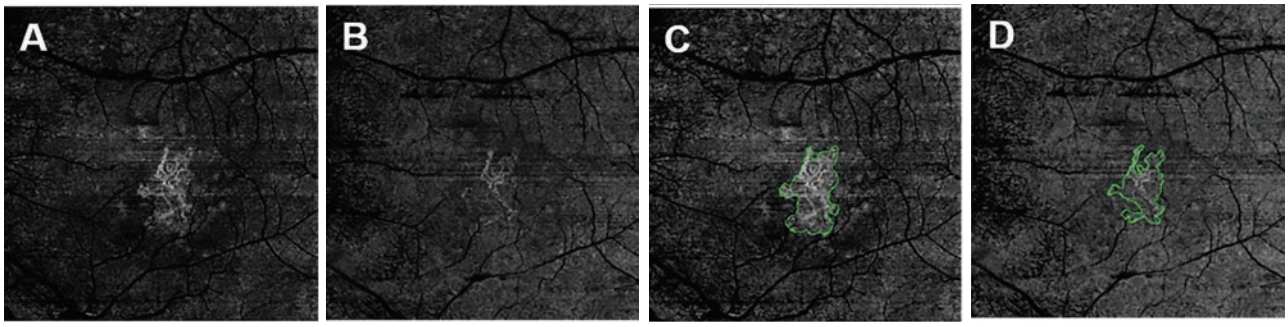


Figure 2. These en face swept-source (SS) and spectral-domain (SD) optical coherence tomography angiography (OCTA) images are of the same eye of the same patient in Figure 1. They were processed from the corresponding volumetric datasets using the same algorithms that were applied to a slab from the outer retina to the choriocapillaris with removal of the projection artifacts from the retinal vasculature. They are: SS-OCTA 6-mm × 6-mm scan (A); SD-OCTA 6-mm × 6-mm scan (B); SS-OCTA 6-mm × 6-mm scan with an outline of the choroidal neovascularization (CNV) and an area of 1.208 mm² (C); and SD-OCTA 6-mm × 6-mm scan with an outline of the CNV and an area of 0.841 mm² (D).

tection of type 1 MNV compared with SD-OCTA imaging.

SD-OCTA vs. SS-OCTA In Detecting CNV

While reports have shown that SD-OCTA imaging detects some MNV in neovascular AMD, the ability of SD-OCTA to detect neovascularization under the RPE appears to have its limitations. This has been reported in cases when choroidal neovascularization (CNV) is known to be present from dye-based angiographic imaging and OCT structural alterations, but SD-OCTA did not detect the neovascular lesion.

A recent report compared the ability of SD-OCTA and SS-OCTA to detect CNV and concluded that SS-OCTA imaging appeared to be better than SD-OCTA in demarcating the full extent of the type 1 MNV.[3]

Another group performed a similar comparison study and reported that SS-OCTA showed more CNV lesions with a higher sensitivity for detecting CNV than SD-OCTA.[4] However, this study had a serious limitation: It used different segmentation strategies to visualize the full extent of the neovascular lesions in the two imaging techniques.

We recently performed a study using a commercially available SD-OCTA instrument and a prototype SS-OCTA instrument (Carl Zeiss Meditec, Dublin, Calif.).[5] We analyzed output data from both instruments using the same segmentation slabs, the same complex decorrelation algorithm, known as optical microangiograhy (OMAG), and the same retinal vascular projection artifact removal algorithm. We showed that SS-OCTA tended to capture a larger area of the neovascular lesions than SD-OCTA in both 3-mm x 3-mm scans and 6-mm x 6-mm scans.

The difference in lesion areas was much larger for the 6-mm x 6-mm scans than the 3-mm x 3-mm scans, which may be due to lower contrast-to-noise ratio (CNR) values in the SD-OCTA 6-mm x 6-mm scans. Later this finding was confirmed by using an automated quantification algorithm to measure the area of MNV.[6]

These results have great importance for the clinician when longitudinally following MNV because the lesions routinely grow larger than the 3-mm x 3-mm scan area, and larger scan areas are needed to visualize the full extent of the MNV.

Comparing Scans With The Same Algorithms

An 81-year-old male followed for neovascular AMD was imaged on both SS-OCTA and SD-OCTA. All images were processed from the corresponding volumetric datasets using the same algorithms that were applied to a slab that extended from the outer retina to the choriocapillaris and included the removal of the projection

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artifacts from the retinal vasculature.

The images from SS-OCTA (*Figures 1A, C, page 11*) and SD-OCTA (*Figures 1B, D*) had the similar size of MNV on 3-mm x 3-mm scans, but the SS-OCTA image showed more details and contrast of the MNV, especially on the lesion border. SS-OCTA images showed larger lesion areas in 6-mm x 6-mm scans and more of the vascular details (*Figure 2*).

When imaged with SS-OCTA, the areas of the MNV tended to be larger than with SD-OCTA, and the differences in the area measurements occurred at the margins of the MNV where recurrences tend to arise.

For real-world imaging of MNV, the clinician will become dependent on OCTA images, not only for the detection of these lesions, but, more importantly, for their follow-up. We predict that we will need scan areas of at least 6-mm x 6-mm to detect MNV growth when deciding whether to treat, observe or extend a treatment interval. RS

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A Twist on Ocular Trauma

(Continued from page 10)

A variety of retinal findings have been reported in patients with DMD. One report described a vascular retinopathy, which may include capillary dropout, saccular venular aneurysms, neovascularization and vitreous hemorrhage.[7]

Additionally, patients with DMD have been found to have electroretinogram (ERG) abnormalities, including a reduced ß-wave or even a negative waveform, similar to congenital stationary night blindness.

Patients with DMD, however, do not report nyctalopia.[5] This ERG response is believed to arise from abnormal neurotransmission between photoreceptors and optic nerve-bipolar cells due to the localization of dystrophin to the outer plexiform layer and its presumed role in neurotransmission.[8]

Some patients with DMD have also exhibited a pigmentary retinopathy, but there is debate in the literature whether these conditions are associated. Prior to his injury, our patient was reportedly free of any overt retinal disease, and we do not believe that his DMD predisposed him to the Purtscher-like retinopathy he developed. RS

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Quotable

Some patients with Duchenne muscular dystrophy have also exhibited a pigmentary retinopathy, but there is debate in the literature whether these conditions are associated. We do not believe DMD predisposed our patient to Purtscher-like retinopathy

UW Medicine EYE INSTITUTE

Dr. Olmos de Koo is an associate professor of ophthalmology and director of the retina fellowship program at the University of Washington, Seattle. Dr. Sweeney is a third-year ophthalmology resident at the University of Washington.

By Paul Hahn, MD, PhD »

Tic-Tac-Toe to Solve SO Migration

A modified, minimalistic technique for silicone oil retention sutures.
With John D. Pitcher III, MD

As surgeons, we sometimes wish all cases were straightforward. But it's the complicated ones that keep things interesting—as long as we have the techniques in our armamentarium to deal with these challenges.

Silicone oil (SO) may be used in complex retinal detachment surgery, but management of SO in patients with aphakia and iris loss or traumatic mydriasis is challenging, because SO can migrate into the anterior chamber and cause keratopathy, elevated intraocular pressure and a suboptimal oil fill.

Ronald Gentile, MD, and Dean Eliott, MD, described placement of SO retention sutures in such cases, which provide an artificial barrier to decrease risks associated with a unicameral eye.[1] Others have described variations of this technique, and a recent series confirmed efficacy in prevention of SO-cornea touch.[2]

Step-by-Step Guide

Here, John D. Pitcher III, MD, provides a step-by-step guide with accompanying video to show how to successfully employ this technique.

He has made some slight modifications to avoid compromising the visual axis and to reduce the number of necessary suture passes (View the online video at: bit.ly/RS_VideoPearl_001). Here are the key steps:

1. Perform a conjunctival peritomy, which is required to bury the prolene suture ends.
2. Perform retinal detachment repair as indicated.
3. With the eye under fluid and prior to performing the final fluid-air exchange, pass a 10-0 prolene suture with a 16-mm STC-6 straight needle (Ethicon, Somerville, N.J.) behind the iris starting 1 mm posterior to the limbus, just off center from the visual axis.
4. A 27-gauge needle can be bent at the hub and then inserted, bevel up, on the horizontally opposite side of the eye, also 1 mm posterior to the limbus and off center from the visual axis.
5. "Dock" the STC-6 needle in the lumen of the 27-gauge needle and then slowly pull both simultaneously out of the eye.
6. Using the same needle and suture, repeat steps 3 through 5 going back in the other horizontal direction, parallel to the initial pass, approximately 2 to 3 mm away from the central visual axis.
7. Cut off the needle and tie the two ends of the prolene suture in a 3-1-1-1 knot at a moderate tension. Trim residual suture ends.
8. Repeat steps 3 through 7 in the vertical direction off center from the visual axis, perpendicular to the initial horizontal sutures. The sutures should be "woven" between the first two, which will bring all four sutures into a single plane, strengthening the surface tension effect (*Figure*). The result is a "tic-tac-toe" grid barrier, which should deviate toward the

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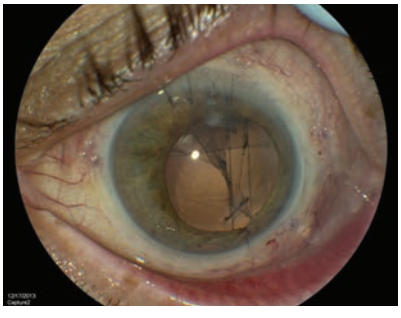

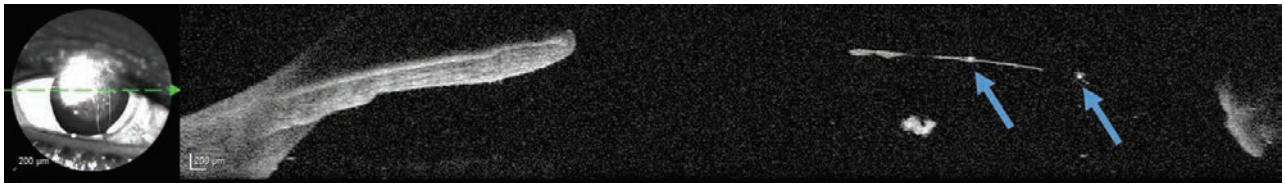

Figure. A 35-year-old man with history of ruptured globe developed a complex retinal detachment with proliferative vitreoretinopathy. Initial repair was complicated by silicone oil (SO) migration into the anterior chamber due to traumatic iris loss and mydriasis. The patient was taken to the operating room where the SO was removed. An inferior iris defect was reconstructed, but traumatic mydriasis resulted in a large pupil particularly superonasally that would not sequester SO. Four retention suture passes deviating toward this superonasal quadrant were woven as a barrier and SO was replaced. Anterior segment optical coherence tomography and anterior segment photographs (below) demonstrate barrier sutures (arrows) in a "tic-tac-toe" pattern with mild fibrin at the interface indicating successful retention of SO posteriorly.

Series: Beyond the Retina

THE CHALLENGE OF MANAGING NAION

How to approach a disease with a variable presentation, complex pathogenesis and unproven treatments.

By Hossein Nazari, MD, Shauna Berry, DO, Ama Sadaka, MD, and Andrew G. Lee, MD

Nonarteritic anterior ischemic optic neuropathy—NAION—can pose a challenge for the retina specialist who may not encounter the disease so frequently. It has a variable visual prognosis, and while multiple medical and surgical treatment options have been proposed and tested, none have been proven. Oral corticosteroids may shorten the duration of optic disc edema and may improve visual outcome, but no randomized, controlled clinical trial has evaluated this treatment.

This article is the first in the series "Beyond the Retina" that aims to update retina specialists on the state of the evidence of non-retinal eye diseases. This review intends to summarize the current evidence-based knowledge in the field of NAION and provide practical clinical recommendations for comprehensive ophthalmologists and retina specialists.

Clinical Presentation

NAION typically presents as acute, unilateral and painless loss of central and/or peripheral vision. Initial visual acuity is variable and may be 20/30 or better in up to 50 percent of cases.[1] Although arteritic anterior ischemic optic neuropathy (AAION) due to giant cell arteritis (GCA) often presents with more severe visual loss, the severity of visual acuity loss is not the sole differentiating feature; both AAION and NAION can produce mild, moderate or severe visual loss. Indeed, about 20 percent of patients with NAION have visual acuity of 20/200 or worse at the time of presentation.[1] Visual field defects in NAION are common and typically consist of nerve-fiber-layer type defects, such as inferior nasal, arcuate or altitudinal defects, although generalized depression, central or diffuse field loss can also occur.[2,3]

Although NAION is typically a disorder of older individuals with vasculopathic risk factors, it may occur at any age, with or without known ischemic risk factors.[1,4] In addition to loss of visual acuity or visual field, patients with NAION may also complain of dyschromatopsia. They usually have a relative afferent pupillary defect (RAPD) in unilateral cases or in bilateral but asymmetric involvement. Slit-lamp biomicroscopy, intraocular pressure and ocular

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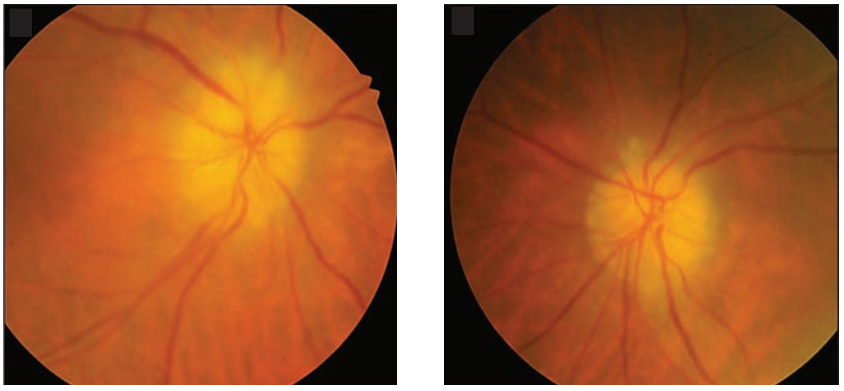
motility testing are normal. In the acute setting, the optic nerve head in NAION shows segmental or diffuse edema with variable degrees of peripapillary superficial nerve fiber layer hemorrhages, mild subretinal fluid or, rarely, cotton wool spots (*Figure*).

The underlying mechanism of NAION is complex and not fully understood. Authors have suggested that NAION occurs due to a transient anterior optic nerve hypoperfusion, particularly at night due to nocturnal hypotension; or it may be secondary to local or systemic disturbances in optic nerve head blood flow, such as arteriosclerosis, vasospasm, hypertension, diabetes, iatrogenic, surgical or non-surgical hypotension or vasoactive medications.[4]

These underlying associated conditions may reduce the autoregulatory capacity of the optic disc and make it more susceptible to perfusion pressure fluctuation. In one study, 52 percent of the patients with NAION noted visual loss upon awakening.[5]

In addition, patients with NAION may be anatomically prone to develop optic disc head perfusion compromise because of a structural risk factor in the optic nerve head, a small cup-to-disc ratio and crowded optic disc head—known as the "structural disc at risk for NAION." A transient ischemic event in a predisposed small, crowded disc may cause ischemic swelling of the axons, leading to compression of capillaries in a restricted optic disc space. This "vicious cycle" of increased swelling causing more compression of the capillaries then inducing further ischemia ultimately results in ischemic infarction of the optic nerve head. Apoptotic loss of ganglion cell bodies and optic atrophy follow the acute ischemic event.



Classic signs of arteritic anterior ischemic optic neuropathy or giant cell arteritis are mild arterial narrowing and chalky white disc swelling in right and left eyes. (*National Library of Medicine: Korean Journal of Ophthalmology*)

Ancillary Tests for NAION

The diagnosis of NAION is primarily clinical; no diagnostic laboratory testing or imaging abnormality exists. We do not generally recommend neuroimaging for typical cases of NAION. Likewise, these typical cases do not require further laboratory testing other than that which helps rule out GCA (*Table 1*). However, atypical cases, such as bilateral, progressive or posterior ION, sometimes require ancillary laboratory and imaging tests.

Optical coherence tomography (OCT) of the optic nerve head can demonstrate detailed morphometric analysis of the optic disc and surrounding retina and retinal nerve fiber layer. Also, optic nerve OCT has documented optic disc elevation, peripapillary nerve fiber layer thickness and peripapillary subretinal fluid.[6,7] Generally, the diagnosis of NAION does not require OCT, but it may be useful in atypical cases or for longitudinal assessment of disc edema and eventual optic atrophy.

Differential Diagnosis

Differentiating AAION (ie, GCA) and NAION is an essential step (*Table 1*). Up to 80 percent of patients with GCA have systemic symptoms and signs that include headaches and periocular pain, jaw claudication, scalp tenderness, abnormal temporal artery, neck pain, anorexia, weight loss, polymyalgia and malaise. However, some patients have occult GCA with none of these signs and symptoms. The presence of any of these indicators should prompt evaluation for GCA in cases of AION.[4]

Transient visual loss or transient diplopia may occur in GCA but should not occur in typical NAION. Therefore, a history of amaurosis fugax prior to AION is suggestive of GCA. Sector or diffuse disc ede-

Take-home Point

Nonarteritic anterior ischemic optic neuropathy—NAION—is the most common form of ischemic optic neuropathy and the second most common optic nerve disease after glaucoma. The clinician's main task in managing NAION is to exclude arteritic AION. The visual prognosis for NAION is variable and visual acuity may partially recover in a large percentage of these patients. This article reviews the existing medical evidence on NAION and what the retina specialist needs to know to differentiate NAION from optic neuritis or papilledema.

Table 1. Differentiating Features Of Acute Unilateral Vision Loss

Feature	Nonarteritic Anterior Ischemic Optic Neuropathy	Arteritic Anterior Ischemic Optic Neuropathy
Age (years)	Typically > 50, but may occur at any age.	Typically > 70, but should be considered in any patient over age 50.
Gender	M=F	F>M
Medical History	Cardiovascular disease, hypertension, diabetes, hypercholesterolemia, sleep apnea, nocturnal hypotension	Polymyalgia rheumatica (up to 40-60 percent)
Associated Symptoms	Typically none	Amaurosis fugax, scalp tenderness, jaw claudication, headache, anorexia, weight loss, fever of unknown origin
Initial Visual Acuity	Variable: 20/20 (30 percent); 20/40 or better (50 percent); 20/200 or worse (20 percent)	Tends to be more severe: 20/40 or better (21 percent); 20/50 to 20/100 (17 percent); 20/200 to count fingers (24 percent); hand motion to no-light-perception (38 percent)
Visual Field	Inferior nasal defect, altitudinal; central vision loss	Variable nerve fiber layer defects, central loss, generalized or diffuse field loss
Relative Afferent Pupillary Defect	Yes	Yes
Erythrocyte Sedimentation Rate/C-Reactive Protein	Within normal limits	Elevated
Optic Nerve, Involved Eye	Hyperemic disc edema, peripapillary splinter hemorrhage	Variable disc edema, pallid edema (chalky white disc); late phase—cupped disc with pale rim
Optic Nerve, Fellow Eye	Small disc with no central cup (the structural "disc at risk" for NAION)	Normal-appearing disc or cupped disc
Other Retinal Findings	Depending on the underlying systemic condition	Choroidal ischemia, ocular ischemia, central retinal artery occlusion, cilioretinal artery occlusion
Fluorescein Angiography	Optic nerve head perfusion delay	Variable delayed choroidal filling
Natural Course	Vision improvement in up to 43 percent	Rare visual improvement
Fellow Eye Involvement	Up to 15 percent	Common fellow eye involvement without treatment
Treatment	Possible use of oral steroids with taper (not proven), control vasculopathic risk factors	High-dose steroids (may require intravenous administration) followed by long-term oral steroids

ma upon examination is common in AION, but the presence of pallid disc edema (ie, pale and swollen disc) in the acute phase is highly suggestive of AAION. Fluorescein angiography might be useful in this setting to evaluate for choroidal perfusion defects that could confirm AAION. Definitive diagnosis of GCA, however, relies upon temporal artery biopsy confirmation.[6]

The differential diagnosis of an acute papillitis other than NAION should consider other causes of optic disc swelling with or without vision loss. In contrast to NAION, optic neuritis commonly presents with vision loss in younger individuals, an RAPD, a central visual field defect, dyschromatopsia and a normal fundus exam—that is, retrobulbar optic neuropathy. The differential diagnosis may also include other infectious or inflammatory causes, such as syphilis or sarcoidosis. Table 2 summarizes the differentiating clinical and radiographic features of NAION compared with optic neuritis and papilledema.

In addition to medications that cause hypotension, NAION has been associated with amiodarone and phosphodiesterase type-5 inhibitors (PDE-5i) that include sildenafil, vardenafil (Levitra, Bayer, Whippany, N.J.) and tadalafil (Cialis, Lilly, Indianapolis).[8] A recent case-crossover study showed that among 43 patients with definite NAION, PDE-5i consumption 30 days prior to an NAION event increased the odds ratio of acute NAION to 2.15 (95 percent confidence interval: 1.06, 4.34).[9] We should advise patients with NAION of the risk these agents pose and that they need to consult their physician regarding continued use.

Visual Outcome

The visual prognosis for NAION is variable; visual acuity may partially recover in up to 43 percent of patients. Resolution of optic disc edema from the onset of visual loss typically occurs within eight weeks.[10,11] Although corticosteroid therapy has been associated with faster resolution of optic disc edema in NAION and may improve visual outcome, no randomized controlled clinical trial

has confirmed this.[10]

The risk of recurrence in the same eye is less than 5 percent, but the fellow eye may be involved in up to 15 percent of patients within five years.[11] In one study, the median interval between NAION in the first eye and occurrence of a new NAION in the fellow eye was 1.2 years, with a range of 16 days to six years.[11]

Management

Multiple medical and surgical treatment options for NAION have been proposed and tested, but none have been proven. Novel treatment methods such as targeting caspase-2 with intravitreal injection of short interfering ribonucleic acid (siRNA)[12] has been tested in a clinical trial.[13] Although a preclinical study has shown the safety of such an approach in test animals, no human study result has been published.[14]

Despite the lack of Level I evidence, retina specialists should discuss the following treatment options and the level of supporting evidence with patients.

• ***Systemic corticosteroid therapy.*** The utility of corticosteroids for NAION is based on the premise that decreased capillary permeability may accentuate the resolution of disc swelling and thus reduce compression of capillaries in the optic nerve head and improve blood flow to the ischemic axons.

A recent large, prospective, nonrandomized, open-label, patient-choice study by Sohan Hayreh, MD, PhD, and M.B. Zimmerman, MD, compared 312 patients who elected treatment with oral prednisone 80 mg/day for 14 days followed by a taper over about two months with 299 controls who elected to remain off corticosteroid.[15]

At six months, 69.8 percent of the eyes with initial visual acuity of 20/70 or worse treated within two weeks of onset had visual acuity improvement compared with 40.5 percent of the untreated eyes (P=0.001). Visual field defects showed improvement in 40.1 percent of the treated group and 24.5 percent of the untreated group (P=0.005).

The authors concluded that steroids were effective in improving visual function compared with the natural history. However, the use of systemic corticosteroids remains controversial due to the conflicting evidence. We tend to discuss the

Table 2. Common Differentiating Characteristics Of Three Major Causes of Optic Nerve Swelling.

Characteristic	Nonarteritic Anterior Ischemic Optic Neuropathy	Optic neuritis	Papilledema
Age (year)	Older, typically >50	Younger, 20-45	Any age, often <35
Gender	M=F	F>M	F>M
Accompanying Disorders	Cardiovascular disease, hypertension, diabetes, obstructive sleep apnea	Multiple sclerosis, neuromyelitis optica	Idiopathic intracranial hypertension, cerebral venous sinus thrombosis, intracranial mass lesions, meningitis
Associated Symptoms	Usually none	Pain with eye movement, abnormal color vision (90 percent)	Headache, nausea, vomiting, transient obscuration of vision, pulse synchronous tinnitus, diplopia
Initial Visual Function	20/40 or better (50 percent); 20/200 or worse (20 percent); variable nerve fiber layer defects	Variable, typically 20/25 or worse, dyschromatopsia common, central or nerve fiber layer defects	Usually normal vision in early cases, enlarged blind spots, nerve fiber layer defects
Relative Afferent Pupillary Defect	Yes	Yes	Typically absent in bilateral and symmetric disease
Optic Nerve, Involved Eye	Sector or diffuse, optic disc edema with superficial hemorrhages and hyperemia	67 percent normal, 33 percent papillitis or disc hyperemia	Bilateral disc swelling (various grades of papilledema)
Unilateral/ Bilateral	Rarely bilateral and simultaneous, but may be bilateral but sequential	10 percent present bilaterally	Almost always bilateral
Imaging Studies	Typically unremarkable	MRI with gadolinium enhancement shows enhancing optic nerve, demyelinating periventricular white matter lesions	Normal MRI and normal MRV in pseudotumor cerebri (PTC), but may show intracranial space-occupying lesion; cerebral venous sinus thrombosis
Lumbar Puncture	Usually unremarkable and LP is generally not recommended for NAION	Variable cerebrospinal fluid (CSF) abnormalities including oligoclonal banding	Elevated opening pressure; may have normal (e.g. PTC) or abnormal CSF contents

potential benefits and risks of high-dose oral corticosteroids with patients who present within two weeks of acute onset, have visual acuity of 20/70 or less and do not have any contraindications for high-dose corticosteroids.

Although intravitreal steroid injection has been proposed for the treatment of NAION, it does not appear to have any advantage over oral prednisone while subjecting patients to the risks of intravitreal injection. However, intravitreal injection may be an option for patients who present with profound visual loss due to NAION and have contraindications for systemic corticosteroids.

• ***Aspirin.*** Two major studies have shown that aspirin has no visual benefit for patients with NAION.[4] However, low-dose aspirin (81 mg) is often recommended to reduce the risk of concomitant cardiovascular events, including possible myocardial infarction or intracranial ischemia.

• ***Neuroprotection.*** The concept of saving the surviving but nonfunctioning "sick" nerve fibers has shown clinical merit in glaucoma and neurologic diseases. Studies have tested multiple agents, including intravitreal erythropoietin and oral levodopa.[16,17]

These studies suffer from a small number of patients and absence of controls. Such studies often do not discuss their results in comparison to the natural history of NAION in which almost 40 percent of patients experience visual recovery. Novel neuroprotection approaches using intravitreal injection of siRNA to target inflammatory cascade mediators have not yet published their clinical results.

• ***Optic nerve sheath decompression (ONSD).*** The Ischemic Optic Neuropathy Decompression Trial (IONDT) is the only randomized clinical trial that evaluated a treatment method for NAION.[3] The proposed mechanism involves reduction of cerebrospinal fluid within the perineural subarachnoid space to resolve potential "compartment syndrome" and possibly improve disc circulation.

The study randomized 258 patients to either follow-up (131) or ONSD (127). The patients assigned to ONSD did no better than patients observed without intervention (32.6 percent gained 3 or more lines of vision vs. 42.7 percent of the observation group). Patients who underwent ONSD had a significantly greater risk of losing vision at six months. Radial optic neurotomy too has been proven futile for NAION.

Little evidence supports the beneficial effects of intravitreal anti-vascular endothelial growth factor (VEGF) agents for NAION.[4] In fact, post-intravitreal injection rise in intraocular pressure and myocardial infarction and stroke have been reported.

In patients with a known precipitating factor such as hypotension, hypovolemia and/or anemia, management should aim to correct the underlying condition immediately. For example, potential treatments for a patient with end-stage renal disease who experiences NAION may include corticosteroid therapy, ambulatory continuous blood pressure monitoring, avoidance of intradialysis sleep and food consumption, lower dialysate solution temperature, blood transfusions and erythropoietin for anemia (and possibly for neuroprotection), antiplatelet therapy such as aspirin and vasopresssors for hypotension episodes.[18]

Because the pathogenesis of NAION differs from intracranial stroke, the proposed treatments for cerebrovascular stroke, including anticoagulation, thrombolytic therapy and antiplatelet treatments, are unproven and are not recommended for NAION.

Conclusion

The clinician's main task in the management of NAION is to rule out AAION. Typical cases of NAION do not require additional evaluation or neuroimaging, but atypical cases should undergo further testing. Differentiating NAION from optic neuritis or papilledema is generally not difficult, but clinicians should recognize the potentially overlapping clinical presentations.

Management should be directed at evaluation and treatment of the underlying vasculopathic risk factors. We recommend consideration of empiric aspirin antiplatelet therapy to reduce cardiovascular event risk in vasculopathic patients—but not to improve visual outcome in NAION.

Some medications, such as PDE5i and amiodarone, may increase the risk of NAION. Although no therapy has been proven effective for NAION, oral corticosteroids may shorten the duration of optic disc edema and potentially improve vision. The prognosis of NAION is variable and unpredictable, but recurrence in the same eye is rare and fellow eye involvement is uncommon. RS

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IS COSOPT THE ANSWER FOR SOME ANTI-VEGF NONRESPONDERS?

Inhibiting aqueous outflow may improve drug retention in the eye.

By Anthony Obeid MD, MPH, and Jason Hsu, MD

Despite the incredible advances in our ability to treat patients with neovascular age-related macular degeneration, we are often frustrated that a subset of patients do not respond optimally to anti-vascular endothelial growth factor agents and continue to have persistent exudation.[1,2] Previous studies have demonstrated that intravitreal drug clearance may be related to outflow from the anterior chamber.[3-6]

Thus, we hypothesized that decreasing the outflow via a potent aqueous suppressant such as dorzolamide hydrochloride-timolol maleate may effectively reduce the clearance of the intravitreal drug.[7] Here, we report on a small study we conducted that aimed to evaluate the effects of adding topical dorzolamide-timolol (Cosopt, Akorn, Lake Forest, Ill.) to a fixed-interval intravitreal anti-VEGF regimen on both the anatomic and functional outcomes in patients with neovascular AMD who had not responded optimally.

We found that in all 10 patients (10 eyes) who completed the study, mean central subfield thickness (CST) decreased from 419.7 µm at enrollment to 334.1 µm at the final visit (P=0.01), and we noted decreases as soon as the first visit after enrollment. We found all had a reduced CST on the final visit after instilling the combination therapy (*Table 1*).

Methodology

Our study recruited patients with neovascular AMD who were incomplete responders to anti-VEGF therapy. We defined incomplete responders as patients who had persistent edema and/or subretinal fluid on spectral-domain optical coherence tomography (SD-OCT) at each of their four prior visits within a six-month period, despite receiving an intravitreal injection of anti-VEGF at each visit. Patients also had to be receiving the same anti-VEGF drug in the study eye at each of these encounters. Table 2 lists the patient characteristics.

The study excluded patients who had a history of uveitis, pars plana vitrectomy, glaucoma surgery, any eye surgery conducted six months prior to enrollment, a history of either anti-glaucoma therapy or sulphonamide allergy, current use of diuretics or corticosteroids and sys-

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Table 1. Anatomical Results After Dorzolamide-Timolol with Anti-VEGF Therapy

Mean Value	At Enrollment	Final Visit	*P* Value
Central Subfield Thickness	419.7 µm	334.1 µm	.01
Maximum Subretinal Fluid Height	126.6 µm	49.5 µm	.02
Maximum Pigment Epithelial Detachment Height	277.4 µm	239.9 µm	.12
Intraocular Pressure	14.5 mmHg	11.9 mmHg	.08

Table 2. Demographics and History of Study Patients

Number of patients	10
Mean age (years)	78.2 (range 65-91)
Number of eyes	10
Mean number of prior injections with same anti-VEGF agent	21.9 (range 7-32)
Intravitreal anti-VEGF agent	
Aflibercept (Eylea, Regeneron)	8
Ranibizumab (Lucentis, Genentech)	2
Treatment interval	
Every 4 weeks	8
Every 5 weeks	1
Every 6 weeks	1

temic contraindications to topical β-blocker therapy. Enrolled patients continued to receive intravitreal injections with the same anti-VEGF drug and fixed-interval regimen that they had been receiving, with the only change being the addition of topical dorzolamide-timolol.

We then instructed the patients to instill the eye drops twice daily in the study eye for the duration of the study period. We assessed their progress by measuring the CST, and gathered additional measurements such as maximum subretinal fluid (SRF) height and maximum pigment epithelial detachment (PED) height for analysis using SD-OCT imaging.

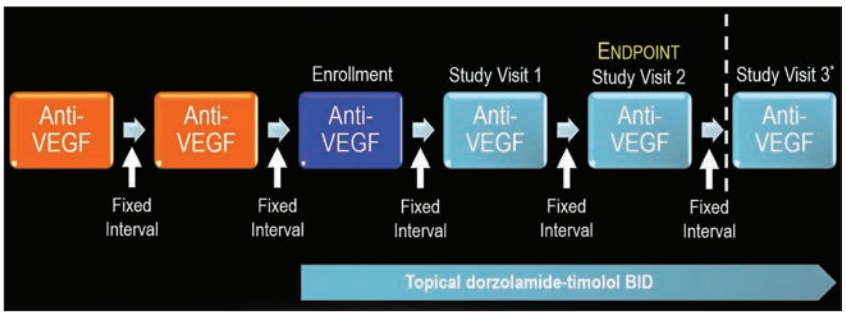

Figure 1. The study design called for patients to be on the same anti-VEGF drug with the same fixed interval between injections for at least two visits prior to enrollment. Following enrollment (dark blue), patients were maintained on the same anti-VEGF drug with the same interval between injections for the study duration. The only addition was topical dorzolamide-timolol BID. While the endpoint was planned at the second visit, eight of 10 eyes stayed on the drops through a third study visit.

At each encounter, we assessed visual acuity (VA) and intraocular pressure (IOP) using the best available Snellen visual acuity and tonometry, respectively. Analysis was subsequently conducted using the paired t-test (GraphPad, GraphPad Software Inc., La Jolla, Calif.) in order to assess the changes in the previously mentioned parameters, with a statistically significant threshold of $P<0.05$. Figure 1 summarizes the study design.

Results

Ten patients (10 study eyes) completed the study protocol and were included for the final analysis. Mean CST decreased from 419.7 µm at enrollment to 334.1 µm at the final visit ($P=0.01$), with a decrease noted as soon as the first visit after enrollment. All patients were found to have a reduced CST on the final visit after instilling the combination therapy.

In addition to the decrease in CST (*Figure 2*), mean maximum SRF height also decreased from 126.6 µm at enrollment to 49.5 µm at the final visit ($P=0.02$) with the decrease also noted at the first visit after enrollment (*Figure 3*). Moreover, all study eyes experienced a decrease in maximum SRF height at the final visit. Additionally, four out of the 10 study eyes demonstrated complete resolution of SRF by the final visit.

Take-home Point

As many as 15 percent of patients with neovascular age-related macular degeneration do not respond to anti-VEGF therapy, which may be due to how the eye clears the agent. The authors performed a small study to explore the hypothesis that dorzolamide-timolol, by suppressing aqueous production and, hence, outflow would improve retention of anti-VEGF agent in the eye. All 10 patients involved showed improvement in anatomical findings after treatment.

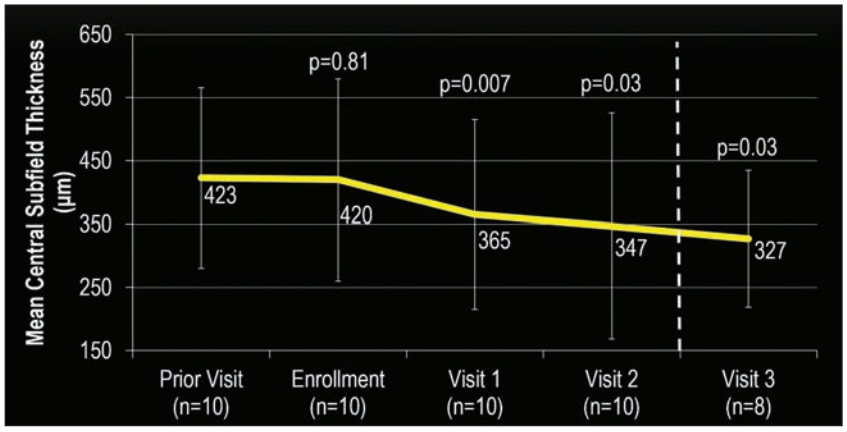

Figure 2. Change in the mean central subfield thickness (CST) after administering anti-VEGF and dorzolamide-timolol in study eyes was fairly stable for the visit prior to enrollment vs. the enrollment visit. However, CST showed a significant decrease as soon as the first visit after starting dorzolamide-timolol that continued throughout the duration of the study.

Mean maximum pigment epithelial detachment (PED) also decreased mildly from 277.4 µm at enrollment to 239.9 µm at the final visit (P=0.12), with five eyes exhibiting a decrease in the maximum PED height at the final visit. Figures 4–6 show the SD-OCT changes in three patients who received the combination therapy. No significant changes in the mean logMAR VA were demonstrable between the first and final visit after enrollment. Mean IOP decreased from the enrollment visit to the final visit from 14.5 to 11.9 mmHg (P=0.08).

How The Eye Clears Drugs

Reports have estimated that 15 percent of patients who suffer from neovascular AMD are classified as non-responders.[1,8] A variety of treatment regimens have been tried in these patients in the past. The combination of intravitreal anti-VEGF and topical dorzolamide-timolol is a novel approach that appears to provide some benefit for such patients. We believe that the therapy works by primarily delaying the outflow of the anti-VEGF drug.[3,7]

While no concrete proof has shown how the eye clears anti-VEGF agents, some evidence has supported the premise that aqueous outflow to an extent removes the drugs.[4,5] In one study, the rate of drug clearance from the vitreous humor paralleled the rate of clearance from the aqueous humor, which reinforces the hypothesis of a common outflow.[6] We already know that dorzolamide-timolol is a powerful aqueous suppressant, decreasing production by up to 50 percent.[7] Perhaps decreasing aqueous production consequently reduces aqueous (and drug) outflow.

One impetus for our study that supports this hypothesis was a study of patients with macular edema due to retinal vein occlusion.[3] In this study, patients receiving a single injection of bevacizumab were randomly assigned to receive dorzolamide-timolol vs. no drops. The results demonstrated a significant difference in the mean central retinal thickness at five weeks, with dorzolamide-timolol-treated eyes having less edema. By nine weeks, there was no difference between the two groups. The study suggested that the aqueous suppressant delayed the clearance of bevacizumab during its duration of action in the early weeks after its administration.

It is also possible that the β-blocker or the carbonic anhydrase inhibitor itself may have led to the positive responses in our study. β-blockers, for example, have been shown to reduce upregulation of angiogenic factors via β2-andregenic receptor blockade[9,10]

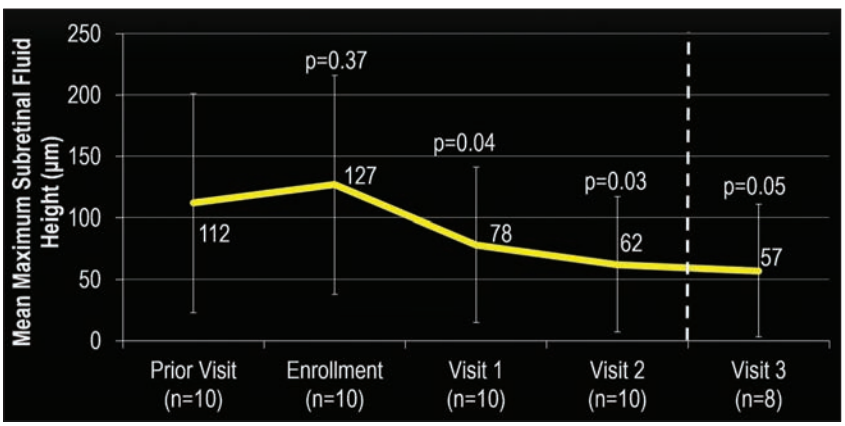

Figure 3. Mean maximum subretinal fluid (SRF) height after administering anti-VEGF and dorzolamide-timolol in study eye was fairly stable in the visit prior to enrollment and the enrollment visit. However, a significant decrease in SRF height was seen as soon as the first visit after starting dorzolamide-timolol that continued throughout the study.

and attenuation of choroidal neovascularization.[11]

One clinical study demonstrated that patients receiving systemic β-blocker therapy were statistically more likely to receive fewer injections of bevacizumab.[12] However, other subsequent studies did not find this same effect, which calls into question whether β-blockers have a true independent effect on choroidal neovascularization.[13,14] It is possible, though, that topical β-blockers may reach a higher intraocular concentration than systemic β-blockers at the usual dose prescribed, which may still lend credence to the hypothetical positive effects of β-blockers on choroidal neovascularization.

Dorzolamide has also been used in the successful treatment of macular edema in a variety of cases.[15-18] The carbonic anhydrase enzyme has been found in both Müller cells and retinal pigment epithelial cells,[19,20] and the inhibition of the enzyme may modulate the pump function in these cells, leading to fluid egress from the retina to the choroid. Dorzolamide has also been shown to increase choroidal perfusion and retinal oxygenation,[21,22] which might suggest an independent role of the drug in reducing macular edema.

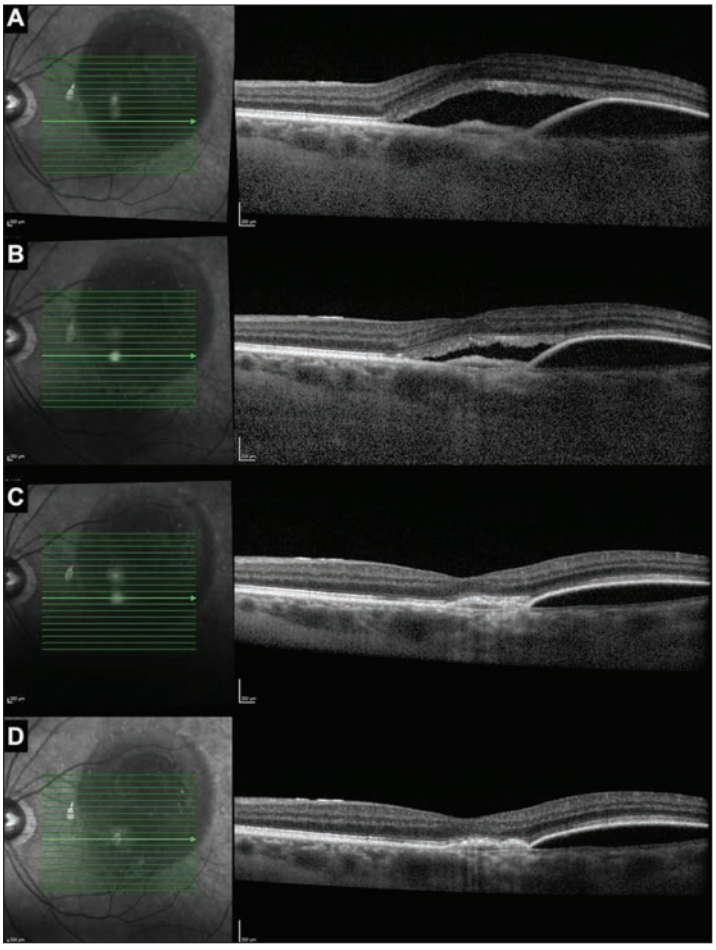

Figure 4. Optical coherence tomography images in one subject at enrollment (A), week four (B), week eight (C) and week 12 (D) show improvement in subretinal fluid and pigment epithelial detachment after the addition of dorzolamide-timolol to the monthly aflibercept regimen. Prior to enrollment, this patient had received 16 aflibercept injections.

Study Limitations

Two clear limitations of our study were the small sample size and the short duration of treatment with dorzolamide-timolol. The short duration may have contributed to the lack of significant visual acuity changes, since visual acuity often lags behind OCT changes in eyes suffering from macular edema.[23-25]

Additionally, the study design does not allow us to determine whether the observed effects were due to the independent action of dorzolamide vs. timolol rather than the combination of dorzolamide-timolol. Another limitation was the lack of a control group and the possibility that our findings represent normal fluctuations of the disease. However, this is unlikely as we only included patients who had persistent exudation over six months prior to inclusion despite fixed-interval anti-VEGF injections. In fact, the mean CST, SRF and PED heights at the visit prior to enrollment and at enrollment were similar, suggesting stability of the persistent exudation. Since we were keeping patients on the same anti-VEGF drug and same fixed-interval that they had been on before enrollment, the likelihood that the improvement noted after addition of dorzolamide-timolol was a mere coincidence seems low.

Patient compliance with the eye drop presented another limitation because the study measured it via self-reporting. However, the consistent drop in intraocular pressure amongst the study eyes suggests, to a certain extent, compliance.

What's Next

The use of dorzolamide-timolol as an adjuvant therapy to intravitreal anti-VEGF agents is a potentially effective regimen in reducing edema and subretinal fluid in eyes that suffer from neovascular AMD and do not respond completely to traditional anti-VEGF treatment. While our study

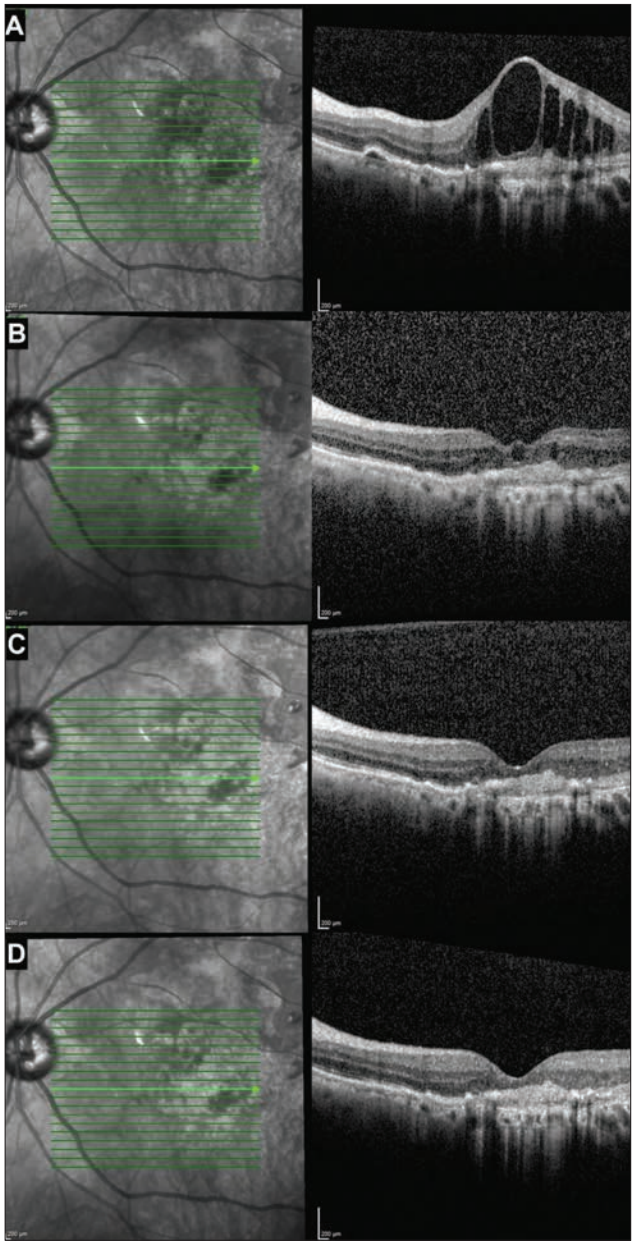

Figure 5. Optical coherence tomography images in a second subject at enrollment (A), week four (B), week eight (C) and week 12 (D) show rapid improvement in the intraretinal edema after the addition of dorzolamide-timolol to the monthly aflibercept regimen. Prior to enrollment, this patient had received 30 aflibercept injections.

did not show any vision changes, the possibility exists that visual acuity may improve with earlier initiation of the drops and longer-term therapy.

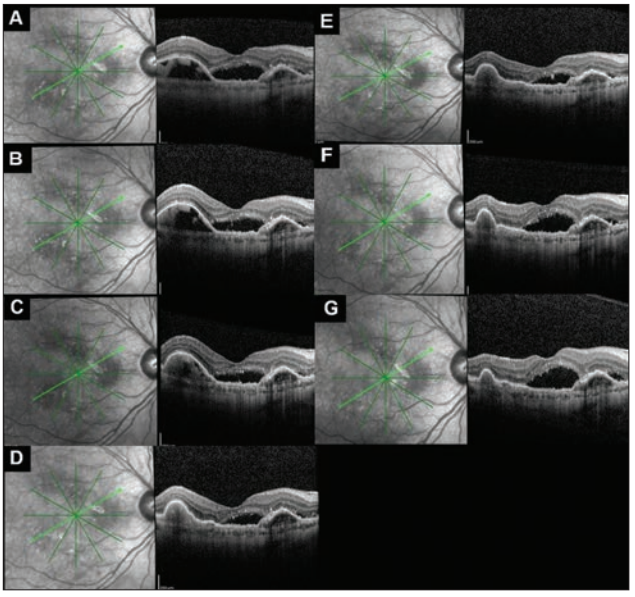

Figure 6. After the addition of dorzolamide-timolol to monthly ranibizumab regimen, optical coherence tomography (OCT) images in another subject at enrollment (A), week four (B), week eight (C), week 12 (D) and week 16 (E) show a gradual decrease in subfoveal subretinal fluid (SRF) as well as pigment epithelial detachment on the left side of the images. Dorzolamide-timolol was discontinued after week 16 and the patient continued to receive monthly intravitreal ranibizumab only. The subsequent OCT images at week 20 (F) and week 24 (G) demonstrate increasing SRF after stopping the drop. Prior to enrollment, this patient had received 30 ranibizumab injections.

Recently, we performed an analysis of the data set from the Comparison of AMD Treatment Trial (CATT)[26] exploring the effect of aqueous suppressants on outcomes. We will present these findings in the near future. A larger, multicenter sham-controlled randomized study with a larger sample size is about to start enrolling; it will seek to determine whether dorzolamide-timolol truly has a beneficial effect on the subset of neovascular AMD patients with persistent exudation despite consistent anti-VEGF injections.

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Beyond Retina: NAION *(Continued from page 19)*

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Tic-Tac-Toe to Solve SO Migration *(Continued from page 14)*

area of sectoral iris loss or traumatic mydriasis.

9. Perform air-fluid exchange and SO injection as usual, taking care to avoid filling oil past the level of the sutures.
10. Close sclerotomies and peritomy with an absorbable suture, ensuring that the conjunctiva covers the prolene sutures.
11. Face-down positioning for the first night can help ensure SO remains posterior.

For cases of sectoral iris loss or traumatic mydriasis, Dr. Pitcher has shown that just four passes with placement deviated toward the area of sectoral loss to spare the visual axis can successfully sequester SO posteriorly. Larger areas of iris loss may require more sutures but the concept remains the same. Techniques such as this to deal with complex situations are important to tilt the odds in your favor for surgical success.

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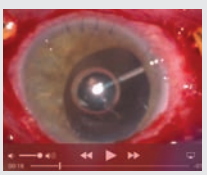

Watch as Dr. Pitcher employs "tic-tac-toe" iris retention sutures to prevent silicone oil migration in a traumatic retinal detachment. Available at: bit.ly/RS_VideoPearl_001.

Gene Therapy and Cutting-Edge Science

PHARMACOGENETICS AND AMD:

WHAT WE KNOW (AND DON'T KNOW) SO FAR

The science is still emerging, but it's reasonable to suspect that genetics can influence response to treatments.

By Parth Shah and Stephen G. Schwartz, MD, MBA

Patients with age-related macular degeneration typically have two avenues of therapeutic intervention—nutritional supplements to reduce the risk of vision loss from intermediate AMD or anti-vascular endothelial growth factor treatment for neovascular AMD. However, the response to either intervention can vary substantially among patients,[1] so a working familiarity of pharmacogenetics—the study of how individual genetic traits may influence the clinical response to medications[2]—can help the retina specialist better understand patients' responses to treatments.[3,4] Genetics may influence some of these responses.

AMD remains the leading cause of irreversible visual loss among the elderly in the United States.[5] The Age-Related Eye Disease Study (AREDS) reported a significant risk reduction among patients with intermediate or advanced AMD who were treated with antioxidants plus zinc,[6] and the potential benefits of anti-VEGF treatments in neovascular AMD have been well documented.[7] This review discusses what we understand about the pharmacogenetics of these approaches to managing AMD.

Complex vs. Monogenic Disease

Diseases with genetic risk factors may be divided broadly into monogenic (single-gene or Mendelian) diseases and complex genetic diseases. These two categories differ in etiology and in general suitability for genetic screening.

Monogenic diseases are typically uncommon. They are, by definition, caused by a single gene defect, or mutation, and follow a recognizable inheritance pattern. In general, individuals with the mutation are likely to develop the disease. Therefore, genetic screening of individuals at risk for monogenic disease is valuable. Examples of monogenic retinal diseases include retinitis pigmentosa, Best vitelliform macular dystrophy and Leber congenital amaurosis. Other articles in this issue discuss the genetics of monogenic disease and the role of screening and testing in managing these diseases.

In contrast, complex genetic diseases may be common. While muta-

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tions cause monogenic diseases, one or more gene variants (risk alleles) plus one or more environmental risk factors have been associated with complex genetic diseases. Risk alleles are not necessarily "abnormal" or deleterious, but are associated with increased risk of disease. This is why they are referred to as *risk alleles* rather than *mutations*. AMD is a complex genetic disease.

Complex genetic diseases do not follow a recognizable inheritance pattern, although patients with these diseases may report a positive family history. The presence of one or more risk alleles does not necessarily imply that the patient will develop the disease. Patients with multiple risk alleles may remain disease-free, while other patients with few or no risk alleles may develop disease. For this reason, genetic screening of individuals at risk for complex genetic disease is less valuable and may provide misleading information compared to screening for monogenic retinal disease.

Complex Profile of AMD

In AMD, 52 risk alleles within 34 loci have been reported to date.[8] The two major associated loci are complement factor H (CFH) (*Figure 1*) and age-related maculopathy susceptibility 2 (ARMS2) (*Figure 2, page 28*);[9] the latter is in very strong linkage disequilibrium with high temperature requirement A serine peptidase 1 (HTRA1)—two loci that cannot be statistically differentiated.[10] Investigators have not determined which of the two is more clinically meaningful. The CFH and ARMS2/HTRA1 variants have been reported to contribute to more than 50 percent of the genetic risk for AMD.[11]

In addition, AMD is significantly associated with multiple non-genetic risk factors, including advancing age, body mass index (BMI) and cigarette smoking.[12] Gene-environment interactions are poorly understood but may also further modulate risk of disease.

When reviewing studies on genotype-phenotype correlations in AMD (or any complex genetic disease), one should remember that a positive finding must be validated in a separate population. Highly significant correlations may be reported that are not clinically "true," especially with large numbers of individual comparisons. This may occur for a variety of reasons, including differences in clinical endpoints, baseline demographics in patient populations and inadvertent selection bias. Therefore, a report of a genotype-phenotype correlation in a single population does not "prove" the association.[13]

Genetic Screening for AMD

At least three genetic tests for AMD are currently available: Macula Risk PGx (ArcticDx, Toronto), Genetic Predisposition Text for Macular Degeneration (EasyDNA, Kent, U.K.) and Asper Ophthalmics (Asper Biotech, Tartu, Estonia). The EasyDNA test is not available to U.S. residents, and a fourth test that had been available, RetnaGene (Sequenom, San Diego) is not currently available.

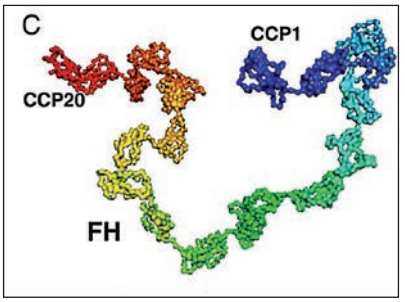

Figure 1. A rendering of the complement factor H (CFH) locus.

A doctor or other provider orders the Macula Risk test, whereas EasyDNA and Asper Biotech are direct-to-consumer genetic tests.

The ACCE model, which the Centers for Disease Control and Prevention and the National Institutes for Health developed, can evaluate the utility of genetic tests.[14] The ACCE model considers four factors:

- **A**nalytic validity—sensitivity and specificity of genetic information.
- **C**linical validity—how well the genetic test predicts the clinical phenotype.
- **C**linical utility—how well the test improves clinical outcomes.
- **E**thical—along with legal and social implications of the test.

A statistic called the area under the curve (AUC), in which an AUC ≥ 0.75 indicates a valid test, has been used to evaluate the accuracy of a test.[15]

The Macula Risk test analyzes 15 variants in 12 loci, plus age, BMI, smoking history and educational level. It has a reported AUC of 0.883 for five-year progression,[15] which indicates excellent analytic and clinical

Take-home Point

Evidence suggests that risk alleles in specific genes may impact development and progression of age-related macular degeneration and possibly response to treatment. While no validated genotype-phenotype associations with either anti-VEGF therapy or Age-Related Eye Disease Study supplementation have been established, AMD pharmacogenetics is an intriguing area of research. However, at this time the evidence is insufficient for using genetic information to guide treatment decisions in individual patients.

validity. However, the clinical utility of Macula Risk is uncertain, because no interventions currently available have been reported to improve outcomes in patients at high risk for advanced AMD. More frequent examination of high-risk patients may seem logical, but to our knowledge there is little or no peer-reviewed evidence that has demonstrated an actual benefit from this approach.

Further, the ethical, legal and social implications of Macula Risk appear substantial. An incorrect false-negative result may give patients a false sense of security, while an incorrect false-positive result could cause anxiety and possibly unnecessary examinations and clinical interventions.

The American Academy of Ophthalmology Task Force on Genetic Testing published recommendations in 2012,[16] which it then updated in 2014,[17] that advise clinicians to offer genetic testing to patients suspected of having a monogenic (Mendelian) disease, and to provide patients with genetic counseling or referral to a genetic counselor. These guidelines also recommended against direct-to-consumer genetic testing and against routine testing for complex genetic diseases such as AMD.

AREDS and Intermediate AMD

With regard to anti-VEGF therapy, many relatively small series and meta-analyses have reported significant associations between various risk alleles and outcomes.[18] However, two large, prospective randomized clinical trials did not validate these findings: Comparison of AMD Treatments Trials (CATT) and Inhibit VEGF in Patients with Age-Related Choroidal Neovascularization (IVAN). The CATT investigators reported no significant associations between variants in CFH, ARMS2, HTRA1 and complement factor 3 (C3) with treatment outcomes.[19] Similarly, the IVAN investigators reported no significant associations between variants in CFH, ARMS2, HTRA1 and others with treatment outcomes.[20]

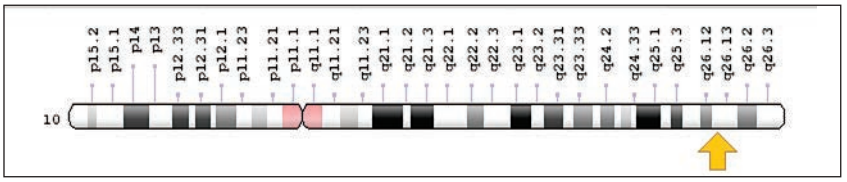

Figure 2. A gene map of the location of the *age-related maculopathy susceptibility 2* (*ARMS2*) loci.

Some risk alleles may be associated with the response to nutritional supplementation in AMD. AREDS was a randomized clinical trial that studied the effect of nutritional supplementation on AMD progression. It randomized a total of 3,640 patients to receive antioxidants (beta-carotene, vitamin C and vitamin E), zinc (defined as zinc plus copper), both, or neither (placebo).[6] The AREDS investigators graded disease severity with fundus photography on a 1-4 scale.

They reported that in patients with category 3 or 4 disease, treatment with antioxidants plus zinc was associated with a significant reduction in disease progression rates by about 25 percent at five years. AREDS defined category 3 disease as at least one large druse (>125 µm), extensive intermediate drusen and/or non-central geographic atrophy (GA). Category 4 disease was defined as central GA, neovascular AMD or visual loss resulting from AMD in one eye.

The AREDS2 study subsequently reported that substituting lutein and zeaxanthin for beta-carotene resulted in similar outcomes,[21] and many clinicians currently recommend the AREDS2 formulation rather than the original AREDS formulation.

Retrospective AREDS Analyses

AREDS obtained and stored genetic data from some participants. Six subsequent retrospective analyses of the original AREDS study data have investigated genetic associations with treatment outcomes (progression rates). These studies have reported conflicting results (*Table*).

Michael Klein, MD, and colleagues investigated 867 patients with category 3 and 4 disease and reported that all patients benefited from antioxidants plus zinc (the AREDS formula). However, significantly more favorable outcomes were associated with patients with no risk alleles at CFH compared to patients with two risk alleles at CFH. There were no associations with ARMS2.[22] The investigators noted this association but did not recommend a change in clinical management.

Carl Awh, MD, and colleagues analyzed 995 patients with category 3 disease and reported a complex relationship in which certain combinations of risk alleles at CFH and ARMS2 were associated with more favorable clinical outcomes with certain nutritional supplements. They concluded that 49 percent of patients in their analysis had risk allele combinations for which nutritional supplements other than the AREDS formula were most beneficial. They suggested that individualized nutritional supplementation based on CFH and ARMS2 variants might

improve clinical outcomes.[23]

The AREDS investigators responded to this publication by performing an "unplanned retrospective analysis" of 1,237 patients with category 3 or 4 disease. This analysis reported that all evaluated patients benefited from antioxidants plus zinc, and that CFH and ARMS2 risk alleles were not significantly associated with treatment outcomes.[24]

Dr. Awh and colleagues published a second study of 989 patients with category 3 and 4 disease. They defined four groups of patients based on CFH and ARMS2 and reported that, for each group, different nutritional supplements were most beneficial.

They concluded that "most" patients with category 3 or 4 disease would have more favorable outcomes if treated with a combination other than antioxidants plus zinc. They noted the lack of an available second population with which to validate their findings, but did recommend individualized nutritional supplementation based on genotype.[25]

The AREDS investigators then attempted to validate these findings by identifying a "residual cohort" of 526 patients from the original AREDS study not included in the subgroup that Dr. Awh and colleagues analyzed. In these 526 patients, the authors found no significant associations between CFH, ARMS2 and treatment outcomes.

Further, to illustrate the importance of a validation sample, the AREDS investigators also analyzed all patients in both cohorts according to astrological sign. They reported that, in the subgroup of patients that Dr. Awh and colleagues analyzed, the signs of Aries and Cancer were associated with significantly worse outcomes when treated with zinc.

Table. Retrospective AREDS Analyses

Study author	N	Year	Key finding
Klein ML, et al.[22]	876	2008	Response to AREDS supplements may be related to *CFH* genotype.
Awh CC, et al.[23]	995	2013	Some combinations of *CFH* and *ARMS2* derived significantly greater benefit from zinc-only supplementation.
Chew EY, et al.[24]	1,237	2014	AREDS supplements reduced rate of AMD progression across all genotype groups; genotypes at the *CFH* and *ARMS2* loci did not statistically significantly alter benefits of AREDS supplements.
Awh CC, et al.[25]	989	2015	Benefit of AREDS formulation seemed to result from a favorable response in only one genotype group, with neutral or unfavorable responses in three other genotype groups.
Chew EY, et al.[26]	526	2015	No significant associations between *CFH, ARMS2* and treatment outcomes.
Seddon JM, et al.[27]	4,124 (eyes)	2016	Effectiveness of antioxidant and zinc supplementation appears to differ by genotype.

However, these associations were not found in the "residual cohort," and were thus not validated.[26]

Most recently, Johanna Seddon, MD, MSc, and colleagues retrospectively studied the AREDS data, analyzing individual eyes (4,124 in total) rather than patients.[27] They reported that antioxidants plus zinc conferred no treatment benefits in patients with two risk alleles at CFH or no risk alleles at ARMS2. The authors did not make any recommendations for clinical management, but did call for additional studies to investigate these outcome differences.

Conclusion

AMD is a complex genetic disease, with both genetic and environmental influences, so analysis of only two risk alleles (CFH and ARMS2) may yield misleading information.[28] No randomized clinical trials have been published regarding genotype-phenotype relationships with AREDS supplementation.

Six retrospective subgroup analyses of the original AREDS data (but none of the AREDS2 data) have been published. The significant associations that some investigators have identified from the AREDS study have not been replicated in a second study population.[12]

Clearly, a prospective randomized clinical trial that was designed to investigate a genotype-phenotype association is preferable to a retrospective subgroup analysis of a randomized trial that was not designed to investigate the association.[29] Unfortunately, no such randomized clinical trial has been published, and it seems unlikely one will occur in the near future. Because of this, clinicians must make treatment decisions based on the available data, which has yielded conflicting results.

In summary, AMD pharmacogenetics is an intriguing area of research, but at this time there is insufficient evidence to use genetic information to guide treatment decisions in individual patients.

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Gene Therapy and Cutting-Edge Science

WHERE GENETIC TESTING FITS IN THE RETINA PRACTICE

Done the right way, gene testing and counseling can help manage patients with inherited retinal diseases.

Karmen M. Trzupek, MS, CGC

Demand for genetic testing for inherited retinal disease has grown in response to a number of advances, most notably the increasing availability and decreasing cost of multi-gene testing, along with the growing availability of gene-based clinical trials. But when should ophthalmologists consider genetic testing? And how can they incorporate that into clinical practice?

The American Academy of Ophthalmology supports genetic testing for all patients with a presumed or suspected inherited, or Mendelian, retinal disease.[1] The AAO Task Force on Genetic Testing states that clinicians should avoid routine genetic testing for genetically complex disorders such as age-related macular degeneration.

The benefits of genetic testing for inherited genetic disease are myriad: A positive genetic test can elucidate or confirm a diagnosis and provide information about likely prognosis. Increasingly, genetic testing is used to screen patients as possible candidates for clinical treatment trials.

Gene-based clinical trials are in progress for some types of Usher syndrome, Leber congenital amaurosis (LCA), retinitis pigmentosa (RP), Stargardt macular dystrophy and choroideremia. Several clinical trials of gene therapy for RPE65-related disease have concluded; the Food and Drug Administration this year could approve the first gene therapy for clinical use.[2,3] (*An article on gene therapy for inherited retinal disease starts on page 35.*)

How Genetic Testing Can Alter Medical Management

In the absence of approved gene-based treatments, though, genetic testing can still change medical management in these meaningful ways (*Diagram, page 32*):

- Reduce the need for additional electrophysiology and/or serological testing clinicians use to narrow differential diagnoses.
- Clarify the need for medical imaging and surveillance associated with syndromic disease.
- Provide guidance in determining ocular surveillance.[1]
- Stratify patients based on risk factors for neovascularization or cystoid macular edema.[4,5]
- Determine when to change medications and supplements, such as when to direct patients with ABCA4-macular dystrophy to avoid supplemental vitamin A.[6]

ABOUT THE AUTHOR

Ms. Trzupek, a certified genetic counselor, leads the ocular genetic counseling team at InformedDNA. She previously worked in the genetics lab at Casey Eye Institute in Portland, Ore., on the team that identified the first gene associated with susceptibility to age-related macular degeneration.

DISCLOSURE: Ms. Trzupek is an employee of InformedDNA and a consultant to Spark Therapeutics.

Testing Clarifies Inheritance

Of course, successful genetic testing also enables accurate counseling regarding inheritance and recurrence risks. The inheritance of RP in particular can be difficult to predict based on family history alone. The Inherited Retinal Disease Clinic at the University of Michigan performed a retrospective study of patients diagnosed with RP, comparing the presumed inheritance type derived from clinical diagnosis and family history with the results of genetic testing. That study, which collected a detailed family history going back three or four generations for every patient, reported that genetic testing identified a different inheritance pattern than pedigree analysis alone in 10 percent of patients.[7]

These discrepancies in underlying inheritance can result from several genetic factors, including *de novo* dominant mutations, X-linked disease in families with manifesting female carriers, dominant disease with reduced penetrance and pseudodominance.[7] These complex genetic factors, often dismissed as too rare for consideration in the typical practice, collectively account for a significant percentage of the underlying genetics of inherited retinal disease.

Genetic Complexity

In 1984, Shom Shanker Bhattacharya, PhD, and colleagues mapped the RP2 gene associated with X-linked RP.[8] The ensuing 30-plus years have seen tremendous advances. Today, nearly 300 inherited retinal disease genes have been mapped, and more than 250 of those genes have been cloned.[9]

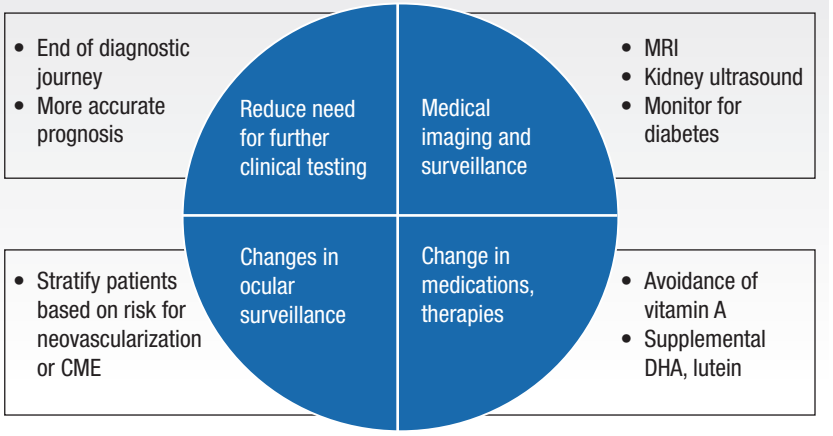
Retinal disease genetics have proven to be a model of genetic complexity. They display not only allelic heterogeneity, where many different disease-causing mutations are found within a particular gene, but also:

- *Genetic heterogeneity*—that is, mutations in different genes may cause the same disease.
- *Phenotypic heterogeneity*—different mutations within the same gene may produce different clinical phenotypes.
- *Clinical heterogeneity*—the same mutation in different individuals, even within the same family, may produce different clinical consequences.

RP is particularly complex. RP can be inherited as an autosomal dominant, autosomal recessive, X-linked or mitochondrial condition. Rare digenic forms have even been documented. While inherited retinal diseases frequently occur as an isolated condition, they may also occur as a presenting symptom of a larger, recognizable genetic syndrome or metabolic disease.

For example, some children with LCA, presenting in infancy or early childhood with apparently isolated retinal disease, may actually have an underlying diagnosis of a syndromic condition such as Joubert syndrome. Prior to multi-gene testing, young children with LCA typically had an MRI to rule out brain abnormalities associated with some of the syndromic forms of LCA.[10] Today, clinicians typically use genetic testing to guide surveillance. Given known genotype-phenotype data, some patients will benefit from MRI, renal ultrasound, hearing evaluations or diabetes monitoring; others will not.

How Genetic Testing Can Influence Changes In Medical Management



Take-home Point

For decades, patients diagnosed with an inherited retinal disease faced the prospect of certain progressive vision loss. Because these conditions were almost universally untreatable, many patients and families lost the motivation to return to the clinic for routine follow-up visits. With the advent of clinical trials, patients are coming back. They are interested in molecular advancements and are frequently eager to undergo genetic testing. Retina specialists who diagnose and manage these patients need a plan for incorporating genetic testing into their care.

Conversely, some genes historically associated with syndromic forms of retinal dystrophy can in fact cause isolated RP or LCA (*Table*). This has significant implications for the interpretation of genetic testing data. While Ush2A is the gene most commonly known to be associated with Usher syndrome type 2, it is also now known to be the most common cause of nonsyndromic RP.[11]

Table. Common Molecular Causes Of Retinitis Pigmentosa[21]

Gene Type	% of Retinitis Pigmentosa Attributed To Pathogenic Variants in the Gene*
ADRP Gene	
RHO	20-30%
PRPF31	5-10%
PRPH2	5-10%
RP1	3-4%
ARRP Gene	
Ush2A	10-15%
ABCA4	2-5%
PDE6B	2-5%
RPE65	~2%
XLRP Gene	
RPGR	70-90%
RP2	10-20%

* U.S. estimates. Estimates vary widely based on ethnicity.

The CLN3 gene has long been known to be associated with neuronal ceroid lipofuscinosis (NCL, or Batten disease), a universally fatal disease that typically begins with childhood onset of severe retinal dystrophy, followed by rapid psychomotor deterioration. Recently though, mutations in CLN3 have been described in multiple unrelated families with nonsyndromic RP.[12] Genes previously believed to cause a very narrow syndromic phenotype may in fact be associated with a high degree of variability in symptom type, onset, severity and progression.[13]

Due to the wide range of phenotypic and clinical heterogeneity known to occur in some genetic diseases—including, but not limited to inherited retinal diseases—a significant shift is underway in medical genetics to begin naming diseases according to their underlying molecular cause. Mutations in RPE65 are known to be associated with not only LCA but also severe early childhood onset retinal dystrophy and juvenile onset RP.[14,15] CRB1 mutations have been associated with LCA, RP, cone-rod dystrophy and RP with preserved para-arteriolar retinal pigment epithelium. Gene-based treatments should benefit patients with underlying mutations in those genes irrespective of their narrowly defined clinical diagnosis.

Laboratory Genetics

For more than 20 years, genetic testing was performed almost exclusively on a single-gene or targeted genetic mutation basis. To date, most patients have been tested for only a few genes that the ordering physician determines are the most statistically likely causative genes. With the advent and application of next-generation sequencing technology, and in some labs whole-exome sequencing, clinicians now routinely order testing for more than 150 genes in a single test.[12,16] As a result, the likelihood of a positive test has risen dramatically. A causative mutation can be identified in 60 to 80 percent of patients with a clinical diagnosis of RP.[16,17]

As is frequently the case, though, more is not always better. With additional genetic testing comes greater complexity of results. Today, in nearly every patient tested for inherited retinal disease, panel testing identifies and reports multiple "variants of uncertain significance." With time, the vast majority of these identified variants will be reclassified as normal genetic variation, but currently they often generate confusion for the patient and create a dilemma for the physician who doesn't have the time to verify and explain those findings. For this reason (as well as cost), the recommendations of the AAO Task Force on Genetic Testing specifically state that clinicians should avoid unnecessary parallel testing and "order the most specific test(s) available given the patient's clinical findings."[18]

What is "unnecessary parallel" testing? The 2016 Recommendations on Clinical Assessment of Patients with Inherited Retinal Degeneration state: "Multi-gene testing is typically necessary for the successful molecular diagnosis of a disease such as retinitis pigmentosa, where >100 causative genes are known."[18]

In some situations, though, large-panel testing is unnecessary, at least as a first-line test. X-linked retinoschisis and Best macular dystrophy are examples of conditions where testing of a single gene is often sufficient to confirm a patient's clinical diagnosis. For achromatopsia, for which testing of a small number of genes will identify the cause of disease in more than 80 percent of cases, a small panel typically suffices.

Importantly, physicians should utilize testing that follows standard guidelines for the interpretation and disclosure of genetic variants. In 2015, the American College of Medical Genetics and the Associa-

tion for Molecular Pathology published a joint consensus document with guidelines for the interpretation of sequence variants.[19] Those guidelines provide a framework for greater standardization of the interpretation and use of variants identified in genetic testing.

Who Should Order and Interpret Genetic Tests?

The AAO task force states: "Ophthalmologists who order genetic tests should either provide genetic counseling to their patients themselves, if qualified to do so, or ensure that counseling is provided by a trained individual such as a board-certified medical geneticist or genetic counselor."

However, this recommendation has met with some controversy. Some professionals believe that only medical geneticists or ophthalmologists with board certification in medical genetics should order genetic testing for inherited retinal disease patients.[20]

Others disagree, stating that there are not enough board-certified medical geneticists to handle the patient volume, and retina specialists already diagnose and counsel patients with inherited retinal diseases using methods other than genetic testing, such as electrophysiology. In addition, as the availability of gene-based treatments increases, a larger number of retina specialists will want to effectively screen their own patients.

Even motivated retina specialists, though, eager to screen patients as candidates for gene-based treatments, struggle to meet the demands of incorporating genetic testing into their medical practices. Many physicians bridge this need by partnering with genetic counselors, who can collect detailed family histories, provide genetic and prognostic counseling, and review molecular testing options with patients. Genetic counselors carefully review genetic variants in test reports and research their potential contribution to disease. Following genetic testing, the genetic counselor may coordinate additional testing of family members, enroll the patient in research or patient registries and discuss clinical trials.

Patients and providers agree that genetic testing can have a multitude of benefits. Besides the clear benefits to patient management, patients and families often describe a peace of mind that comes at the end of a sometimes long diagnostic odyssey. They frequently feel more engaged in the disease community and more empowered to participate in (and comply with) their health care.

But psychosocial risks to genetic testing exist as well. Patients sometimes describe an emotional burden from learning that their family members may carry a genetic disease. Others experience intense disappointment when genetic testing does not indicate that they will qualify for a clinical trial. Younger patients may struggle with family planning decisions.

For all of these reasons, patients receiving genetic testing results deserve the time of a trained professional who can explain the findings and identify individuals in need of additional support.

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Gene Therapy and Cutting-Edge Science

GENE THERAPY:
THE NEW FRONTIER FOR INHERITED RETINAL DISEASE

An update on clinical trials of gene replacement therapy for retinal dystrophies.

Christine Kay, MD

In the past 15 years, research in the field of retinal gene therapy has exploded. While no treatments have yet been approved for any inherited retinal dystrophies, clinical trials involving retinal gene therapy are creating hope for future therapies for afflicted patients. Consequently, retina specialists must now be able to appropriately diagnose counsel patients with retinal dystrophies who may be candidates for clinical trials.

This article will focus on updates in retinal gene therapy with an introduction to viral-based gene therapy, followed by a discussion of current retinal gene therapy clinical trials. The goal is to give the retina specialist a framework for evaluating and counseling these patients as they come through our clinics.

Inherited Retinal Disease: A Brief Review

Inherited retinal diseases can be categorized by anatomic location in the eye—the macula, fovea, choroid or vitreous. Some diseases are more diffuse and affect all photoreceptors in the retina with varying degrees of insult to either rods or cones.

We categorize these diffuse diseases into two broad categories: stationary or progressive. Stationary diseases are typically early onset, such as congenital stationary night blindness, whereas progressive diseases tend to be of later onset, such as retinitis pigmentosa (RP). Other inherited retinal diseases are part of larger syndromes or associated with systemic disease (*Table 1, page 36*).

Multiple clinical trials are ongoing for many of the diseases listed in Table 1. RP, the most common retinal dystrophy, has a prevalence of roughly 1:4,000.[1] RP associated with the MERTK gene (for MER proto-oncogene tyrosine kinase) is an autosomal recessive form of the disease that is the subject of a retinal gene therapy clinical trial.[2]

Stargardt disease is another common retinal dystrophy (prevalence: roughly 1:8,000)[3] that is the focus of multiple clinical trials, including a subretinal lentivirus gene therapy trial,[4] a stem cell therapy trial[5] and an oral drug trial.[6] Less prevalent diseases, including Leber congenital amaurosis (LCA), achromatosopia, X-linked retinoschisis (XLRS), Usher syndrome and choroideremia, are all subjects of current gene therapy clinical trials. Given these clinical trials, the need for accurate diagnosis and counseling has substantially increased.

A typical examination of a retinal dystrophy patient starts with a detailed history, with a particular focus on family history, followed by a comprehensive ophthalmologic exam. Imaging—particularly optical coherence tomography, fundus photography and autofluorescence—electrophysiologic testing and visual field testing can also play an important role in the evaluation.

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DISCLOSURES: Dr. Kay disclosed receiving clinical trial funding from AGTC, Foundation Fighting Blindness, Alkeus and QLT, and serving as a consultant to AGTC and Second Sight.

Table 1. Inherited Retinal Diseases

Macular Dystrophies
- Best Disease
- North Carolina Macular Dystrophy
- Sorsby Macular Dystrophy
- Stargardt Disease
- Pattern Dystrophy
- Malattia Levintinese (Doyne's Honeycomb)

Foveal Dystrophies
- X-linked Retinoschisis
- Albinism
- Isolated Foveal Hypoplasia

Choroidopathies
- Choroideremia
- Gyrate Atrophy

Vitreoretinopathies
- Stickler Syndrome
- Wagner Syndrome
- Familial Exudative Vitreoretinopathy
- Norrie Disease
- Autosomal Dominant Neovascular Inflammatory Vitreoretinopathy

DIFFUSE PHOTORECEPTOR DISEASES

Stationary/Congenital Onset
- Congenital Stationary Night Blindness
- Leber Congenital Amaurosis
- Achromatopsia
- Blue Cone Monochromacy
- Enhanced S-Cone/Goldmann-Favre Syndrome

Progressive/Later Onset
- Retinitis Pigmentosa
- Cone Dystrophy
- Cone-Rod/Rod-Cone Dystrophy

Syndromic
- Usher Syndrome
- Bardet-Biedl Syndrome
- Kearns-Sayer Syndrome
- Ahlström's Syndrome

Systemic/Metabolic
- Refsum Disease
- Bassen-Kornzweig Syndrome (abetalipoproteinemia)
- Batten Disease

A common misconception about inherited retinal disease is that the lack of a family history argues against a genetic origin of disease. The majority of inherited retinal diseases are passed on in an autosomal recessive pattern, and often the "proband" (or affected individual) is the only reported person in a large family pedigree. Children of carriers of a recessive disease have only a one-fourth chance of having the two mutated alleles.

Similarly, the likelihood for a patient with autosomal recessive disease to pass the disease to offspring is remarkably low if the other parent is unaffected, and the prevalence of the carrier state of most retinal dystrophy mutations is quite low in the general population. Obviously, consanguinity can markedly increase the likelihood of seeing recessive disease manifest—a phenomenon known as pseudo-dominance. Eliciting this history in the clinical examination can help us better predict the inheritance pattern.

Once we establish a clinical diagnosis and an inheritance pattern, we may offer genetic testing for confirmation of disease.

Genetic Testing for Retinal Dystrophies

The key to developing possible gene-based therapies is efficient and accurate genotyping. Gene therapy is effective only when the genetic defect is identified in a given inherited retinal dystrophy. In the past 35 years, more than 200 retinal dystrophy genes have been identified and another 50 have been mapped—that is, the chromosomal location is known but the gene has not been identified (*Figure 1*).

Research-based or commercially available testing has its pluses and minuses. Typically, research-based testing can be at least partially funded by grants, resulting in lower patient cost. However, not all patients are candidates for grant-funded genetic testing options and results typically take much longer to receive.

Commercial labs often offer several-week turnaround and relatively comprehensive retinal dystrophy panels or exome sequencing, but patients' out-of-pocket costs can be high depending on insurance coverage.

Take-home Point

Multiple ongoing retinal gene therapy trials are underway, and one genetic treatment for Leber congenital amaurosis has been submitted for regulatory approval. The most common form of gene therapy involves replacing a protein that the cell no longer expresses because of a genetic mutation—known as replacement gene therapy. Most of the ongoing retinal gene therapy trials utilize subretinal delivery of the vector that carries new genetic material. This article describes existing techniques for delivering viral vectors to affected cells and reviews the clinical trials of genetic therapies and the congenital retinal dystrophies they are targeting.

Many academic centers with ocular genetics services collaborate with genetic counselors. Outside of academic centers, some retina specialists who specialize in inherited retinal dystrophies will coordinate genetic testing and offer patient counseling. A service like InformedDNA can offer ocular genetic counselors via telemedicine.

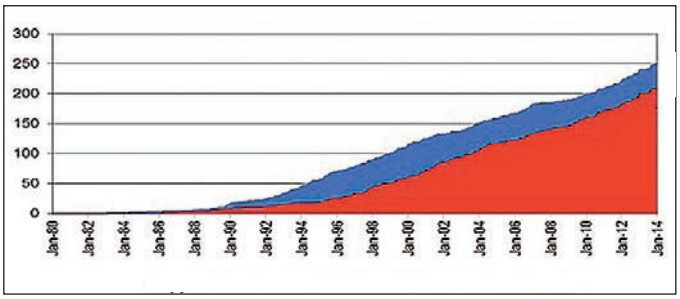

Figure 1. The number of retinal dystrophy genes mapped (blue) and identified (red) from 1980 to 2014 has increased steadily, and new retinal dystrophy genes continue to be discovered. (*Courtesy Stephen P. Daiger, PhD. Used with permission*)

Gene Therapy 101

Gene therapy involves use of a vector to carry the gene of interest into the host cell. Bare DNA, nanoparticles or viruses are examples of vectors, with viruses the most commonly used in clinical gene therapy (*Figure 2*). Existing techniques for viral vector delivery involve intravitreal and subretinal administration (*Figure 3, page 38*). Future techniques may include suprachoroidal and sub-internal limiting membrane techniques.

Once a viral vector is inside the nucleus, the host cell machinery can mediate the gene expression and translation into a protein product.

Adeno-associated virus (AAV) is particularly well suited for gene therapy because it is nonpathogenic, nonimmunogenic and episomal. That is, it does not integrate into the host DNA, but rather remains separate inside the nucleus where it is effectively expressed and translated into protein.

One limitation of AAV is its packaging size; this vector can only hold a 4.7-kb transgene. Scientists have taken advantage of the ability of AAV to encapsulate and deliver DNA into human cells by manipulating the virus genome to remove genes that cause disease and insert therapeutic ones. To create an AAV vector carrying a transgene of interest, the transgene is co-transfected with the rep (or replication) and cap (or capsid) viral DNA into a packaging cell, along with helper adenovirus required for replication.

Once the helper adenovirus is eliminated, the end product is the transgene of interest carried inside a viral capsid. AAV capsids can be modified (by introducing point mutations in the viral capsid genome) to make them more efficient at transduction.

The SAR422459 trial[4] (previously known as StarGen) and UshStat trials[8] are using lentivirus as the vector. UshStat is a gene therapy developed by Sanofi for Usher Syndrome type 1B (USHB1).[9]

Lentivirus is a subclass of retrovirus in which viral genome in the

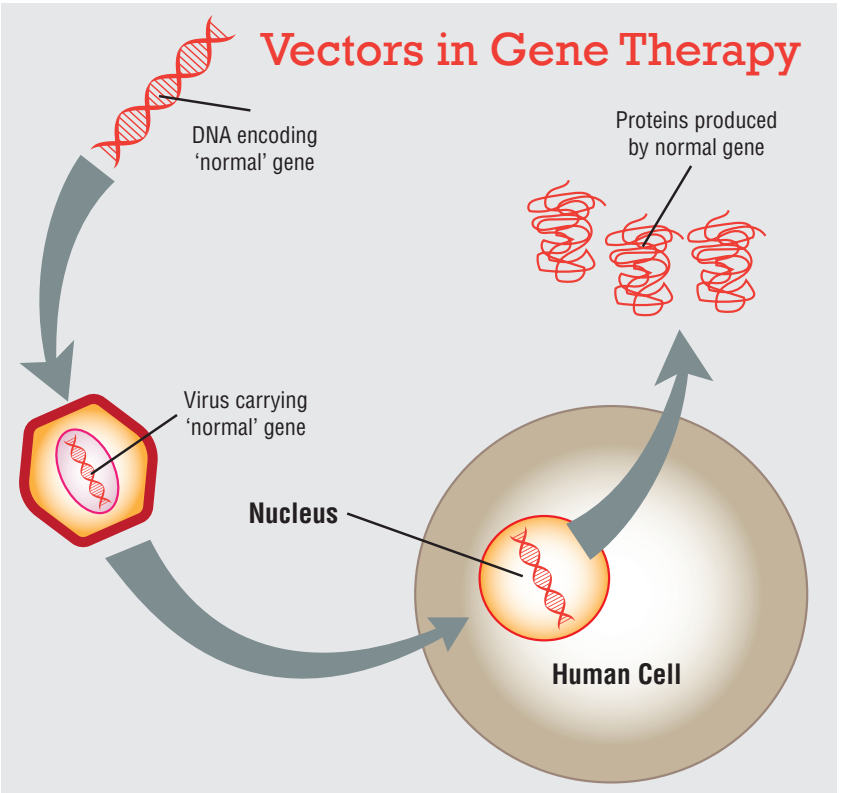

Figure 2. In gene therapy, a viral vector carries the gene of interest into the host cell. Once this viral vector penetrates the nucleus of the host cell, the host cell machinery can mediate expression and translation of the gene into a protein.

form of RNA is reverse-transcribed when the virus enters the cell to produce DNA. Lentivirus is believed to integrate into the genome and can infect both dividing and non-dividing cells. Lentivirus has a much larger carrying capacity than AAV (packaging capacity of 8 to 10 kb), making it the ideal vector for treating retinal genetic disorders with larger affected genes (such as the ABCA4 gene implicated in Stargardt disease).

"Replacement" gene therapy is the most common clinically relevant gene therapy. It involves replacing a protein that a cell no longer expresses due to a genetic mutation in an autosomal recessive condition. This article will focus on replacement gene therapy.

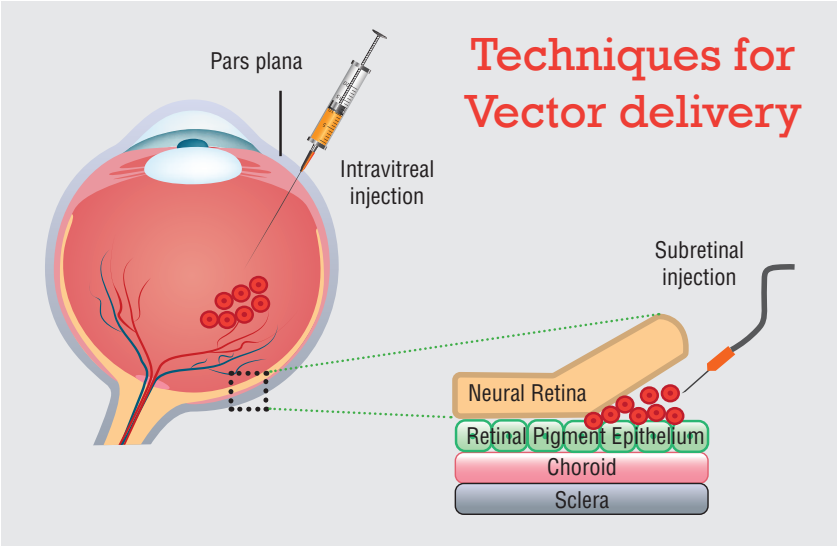

Figure 3. Intravitreal and subretinal administration are the existing techniques for viral vector delivery. With subretinal delivery, investigators have raised concerns about the mechanical trauma that foveal detachment may induce to photoreceptors.

Other Forms of Gene Therapy

Multiple other forms of gene therapy exist. For example, a growth factor can be added in conditions where we do not know the genetic mutation or the conditions are genetically multifactorial. The Oxford Biomedica-sponsored RetinoStat trial for age-related macular degeneration involves expressing endostatin and angiostatin to provide sustained release of an anti-VEGF protein.[7]

Optogenetics (*Box, page 40*) involves genetically altering ganglion cells to become photosensitive. This would be useful in retinal degenerations in which the photoreceptors have already suffered extensive damage. For dominant conditions, we cannot replace a missing gene, so the option of suppression gene therapy arises, which involves small or short interfering RNA (siRNA).

Gene-editing techniques, such as CRISPR (clustered regularly interspaced short palindromic repeats), aim to genetically alter or modify DNA. This has been done *in vitro* and in mouse models, and has been used as a technique for controlling dominant/negative effect.

Surgical Considerations

As we gain experience from human retinal gene therapy clinical trials, we are learning that the mode of gene therapy delivery is an important determinant of both safety and potential efficacy. The majority of retinal gene therapy trials use subretinal delivery of a viral vector to efficiently transduce photoreceptors. Preclinical animal models have reported success with subretinal delivery for transduction efficiency and rescue of the condition, so logic dictates that subretinal induction would follow in human trials.

We do not yet have a viral vector that can efficiently transduce photoreceptors via intravitreal delivery in any of the inherited retinal diseases currently in human gene therapy trials. The XLRS study utilizes an AAV vector and an intravitreal mode of delivery, but given the ubiquitous intraretinal expression of RS1 in this disease, the photoreceptors do not need to be transduced.[10]

The LCA2 studies offered some insight into the importance of localization of the subretinal bleb. In most of the LCA2 Phase I/II trials, subretinal blebs were placed in variable locations; some involved the fovea, others were extramacular. However, some investigators have raised concerns over the mechanical trauma that foveal detachment may induce to photoreceptors.[11] Patients in gene therapy clinical trials are often young, phakic and do not have a posterior vitreous detachment, all of which are certainly considerations in surgical planning.

Instrument selection for creating the subretinal bleb is also important. Options include extendible or non-extendible cannulas, probes of 38 to 41 gauge, and manual vs. automated vector delivery. The surgeon can use the viscous fluid injector system for a foot-pedal automated injection or have a second surgical assistant man-

ually inject the vector via a syringe and tubing system.

Another option is to create a "prebleb" with basic salt solution prior to injection of the vector to minimize the risk of losing the vector into the vitreous. Lifting the retina can take several attempts once the retinotomy is made with the cannula. Intraoperative OCT can help confirm the subretinal location of the vector.

Retinal tissue with degeneration and thinning is more prone to macular hole formation and iatrogenic retinal tears than healthy, thick retinal tissue. Typically surgeons try to not use air or gas to avoid bleb displacement. Postoperative supine positioning can maximize settlement of the bleb over the posterior pole. The timing for blebs to settle after surgery varies depending on the health of the retinal pigment epithelium and other factors.

Clinical Gene Therapy Trials

Active clinical gene replacement trials are targeting Stargardt disease, Usher syndrome, RP, XLRS, choroideremia, achromatopsia and Leber congenital amaurosis 2 (LCA2) (*Table* 2). These trials use either AAV or lentivirus as viral vectors.

Stargardt Degeneration Trials

Stargardt disease is one of the most common inherited retinal dystrophies, with a prevalence of approximately 1:8,000.[3] Typical autosomal recessive Stargardt disease is associated with mutations in ABCA4 gene expressing the photoreceptor-specific ABCA4 protein, a member of the superfamily of ATP-binding cassette (ABC) transporters. Clinically, patients typically develop central visual loss as a result of progressive accumulation of lipofuscin in the RPE with the development of yellowish "pisciform" flecks and eventual macular atrophy.

TABLE 2. Current Gene Therapy Clinical Trials

Phase I/II Trials

- Stargardt: SAR422459 (StarGen)
- Usher 1B: UshStat
- Retinitis Pigmentosa: Optogenetics
- X-linked Retinoschisis: Intravitreal adeno-associated virus-2 (AAV2)
- Choroideremia: AAV2
- CNGB3-associated Achromatopsia: AAV2
- CNGA3-associated Achromatopsia: AAV2
- MERTK-associated RP: AAV2

Phase III Trial

- RPE65-Leber Congenital Amaurosis: AAV2

Depending on the severity of the mutations in the ABCA4 gene, there may be a wide spectrum of phenotypes, ranging from relatively mild and late-onset localized macular disease to earlier-onset diffuse cone-rod disease. A 48-week, Phase I/IIA dose-escalation trial is investigating SAR422459, a lentiviral vector gene therapy carrying the ABCA4 gene formerly known as StarGen, for the treatment of Stargardt disease.[12] Eligible patients must have two pathogenic ABCA4 gene variants confirmed by segregation analysis of parental samples.

This study is investigating vitrectomy with subretinal injection of SAR422459. The primary objective is to assess the safety and tolerability of SAR422459, with the secondary objective to evaluate biological activity. After 48 weeks, patients are encouraged to continue follow-up in a long-term safety study. At this writing, 23 patients have been enrolled, and no significant changes in best-corrected visual acuity have been reported in either the treated or untreated fellow eyes. The plan is to continue enrollment in the cohort of youngest patients with early-onset Stargardt disease and evidence of rapid progression of disease (ages 6 to 26 years; all other cohorts involve patients 18 years or older).[12]

Usher Syndrome

Usher syndrome refers to a clinically and genetically heterogeneous group of autosomal recessive disorders which account for the most frequent cause of combined deafness and blindness in humans, with an estimated prevalence of 3–6:100,000.[13]

Usher syndrome has three clinical subtypes: USH1; USH2; and USH3. The severity and progression of hearing loss and the presence or absence of vestibular dysfunction distinguish these subtypes. USH1 is the most severe form in terms of the onset/extent of hearing loss and RP. The genetic mutation MYO7A (Usher 1B) accounts for approximately 30 to 50 percent of all USH1 cases.[9]

MYO7A (Myosin 7A) encodes an actin-based protein that performs critical motility functions in both the inner ear and retina. Patients with USH1B are born with profound neurosensory deafness, have vestibular dysfunction (that is, they often have a history of delay in walking), and develop early retinal degeneration in childhood.

A trial is investigating SAR421869 (UshStat), a lentiviral gene therapy administered via subretinal injection for the treatment of RP in patients with Usher syndrome type 1B (MYO7A gene defect). All patients must have two confirmed mutations in MYO7A.[14] As of this writing, nine adult patients have been treated.[15] A majority of these patients have shown an initial postoperative drop in BCVA and visual fields that improved to

baseline within two weeks in early unpublished results. The vision was stable (in either the treated or untreated fellow eyes) after 48 weeks in a majority of patients. A separate cohort will provide the opportunity to extend the study to include pediatric patients ages 6 years and up.

X-linked Retinoschisis

XLRS is an X-linked disorder that affects approximately 1:5,000 to 1:20,000 individuals.[16] The disease begins early in childhood, and affected boys typically have BCVA of 20/60 to 20/120 at initial diagnosis. Severe complications such as vitreous hemorrhage or retinal detachment occur in up to 40 percent of patients, especially in older individuals.[16]

The causative gene was identified in 1997 and named retinoschisin 1 (RS1).[15] The gene codes for the retinoschisin protein, which normally provides lateral adhesion that holds retinal cells together. RS1 gene mutations alter the protein to disrupt cell structure. Without normal retinoschisin, the layers of the retina split. Affected individuals typically have early central vision loss and can develop peripheral schisis, exudate or retinal detachment. This damage often forms a "spoke-wheel" pattern in the macula as seen on clinical examination and OCT.

Research has shown that intravitreal AAV delivery can rescue the condition in mice, likely due to the diffuse expression of RS1 throughout the retina as well as the relatively increased retinal permeability that abnormal retinal morphology causes.[18] This is the first replacement gene therapy trial investigating the safety and efficacy of intravitreal gene delivery for an inherited retinal dystrophy.

Ongoing are two Phase I/II studies of an intravitreal-administered AAV-RS1 vector. The National Eye Institute is evaluating three different increasing dose levels of an AAV-RS1 vector in up to 24 adult patients with VA of 20/63 or worse in one eye.[19] In the second study, the biotechnology company AGTC is evaluating an AAV-RS1 vector in up to 27 patients.[10] The latter study involves three initial groups of adult patients receiving increasing dose levels of the vector and will also evaluate the maximum tolerated dose level in patients 6 years and older.

Choroideremia

Choroideremia is an X-linked recessive disorder of a genetic defect in RAB escort protein 1 (REP1) that causes degeneration of RPE and photoreceptors. It can lead to severe and diffuse chorioretinal degeneration. Patients experience gradual vision loss starting from the periphery and advancing toward the fovea. Multiple Phase I and II trials of the AAV.REP1 vector are ongoing at several sites.

In a Phase I/II study, two patients with advanced choroideremia who had low baseline BCVA gained 21 and 11 letters in vision, respectively, despite undergoing retinal detachment.[2] Four other patients with near normal BCVA at baseline recovered to within 1 to 3 letters. Maximum sensitivity measured with dark-adapted microperimetry increased in the treated eyes.

In all patients, the increase in retinal sensitivity over six months in the treated eyes correlated with the vector dose administered per area of surviving retina.[20] The early improvement observed in two of the six patients was sustained at 3.5 years after treatment despite progressive degeneration in the control eyes.[21]

Other trials of subretinal placement of the AAV.REP1 vector are ongoing, including a Phase I/II trial Spark Therapeutics is sponsoring.[22]

Optogenetics, The Newest Frontier

Optogenetics is an innovative new field that involves genetically modifying neurons to express light-sensitive ion channels that may prove beneficial for multiple retinal dystrophies regardless of the specific causative genetic defect.

Additionally, optogenetics may still be useful even when photoreceptors are severely damaged or even missing. Optogenetics targets ganglion cells or inner retinal cells (bipolar cells), obviating the need to target the remaining viable photoreceptor cells. This form of gene therapy opens a new frontier and may offer a broader treatment for a variety of retinal degenerations with a possibly larger window of opportunity for successful intervention in progressive disease.

RetroSense Therapeutics, acquired last year by Allergan, is sponsoring a single-center, dose-escalation study in RP of a photosensitive gene (channelrhodopsin-2) delivered via intravitreal injection.[35] The primary endpoint is safety. The trial will involve 15 adult subjects with advanced RP and visual acuity no better than hand motions.

Achromatopsia

Achromatopsia is an autosomal recessive disease that affects approximately 1:30,000 individuals and is associated with the complete loss of cone function.[23] Achromatopsia is of congenital-onset and relatively stationary, with clinical findings of poor central visual acuity (usually 20/200), nystagmus, severe photophobia and complete loss of color discrimination. On electrophysiology testing, patients have nonrecordable cone-mediated responses.

The two genes most commonly associated with achromatopsia are CNGB3 and CNGA3. A Phase I/II

dose-escalation study sponsored by AGTC evaluating an AAV-CNGB3 subretinal vector in patients with CNGB3 achromatopsia is ongoing at four sites in the United States.[24]

MERTK-RP

The MERTK-associated form of autosomal recessive RP is very rare, with isolated patient populations identified in the Middle East and most recently the Faroe islands.[2] A Phase I clinical trial utilizing an AAV2 vector with an RPE-specific promoter driving MERTK was recently completed in Saudi Arabia.[2] Six patients were treated with subretinal injection of an AAV vector expressing MERTK, without any serious adverse events. Three of these patients displayed measurably improved visual acuity in the treated eye following surgery, although two of them had lost that improvement by two years.

LCA2 (RPE65-associated LCA)

Because of its early onset and the availability of multiple animal models, innovators have focused a tremendous amount of attention on developing a gene-based therapy for RPE65-associated LCA, or LCA2 (prevalence 1:100,000).[25] Multiple Phase I/II trials for RPE65-associated LCA have been either completed or are ongoing. These trials have suggested that improvement in retinal function, as measured by cone and rod sensitivity, is detectable within the first month after treatment[26-28] and may persist at one and three years.[29]

Despite these promising results of early visual gain, reports of visual acuity loss after treatment[30] and continued photoreceptor degeneration at three years have emerged.[31]Although these findings of progressive degeneration are somewhat discouraging, they do provide context for an educated and realistic interpretation of findings from these exciting Phase I/II trials as we move into treatment trials for other inherited retinal disorders.

The recently completed Phase III trial of SPK-RPE65 for treatment of RPE65-associated LCA reported that treated patients displayed improved sensitivity to dim light compared to controls ($P<0.001$) with no significant difference in visual acuity between the two groups.[27] The 31 subjects were randomized 2:1 to an early treatment arm or a one-year treatment-delayed arm.[27] Both eyes received a subretinal injection of 300 µL of AAV, with the second eye treated within 18 days of the first.

The primary endpoint for this trial was mobility testing in an obstacle course with one eye patched. Treated patients scored better than controls ($P<0.001$), meaning that these treated patients could navigate the maze in lower-light conditions. The secondary outcome was full-field light sensitivity, which was done with both eyes open.

The trial reported no serious adverse events. All ocular events were mild. They included transient elevated intraocular pressure in four subjects, cataract formation in three, retinal tears that resolved after laser in two subjects and transient mild eye inflammation in two subjects. Spark Therapeutics has filed a Food and Drug Administration application for approval of this therapy. That could pave the way for future retinal gene therapies and certainly raise awareness of the need for accurate clinical diagnosis of retinal dystrophies and genetic confirmation of disease.[27]

Optimizing Vectors, Delivery

Groups are also continuing to work on optimizing vectors for "potency" to possibly increase the therapeutic effect of gene transfer.[32] Some investigators believe that earlier treatment in these progressive retinal dystrophies may offer the best chance of sustained visual recovery. Phase I/II trials have shown no direct correlation between patient age and treatment response, although they did report less dramatic improvements in retinal sensitivity in younger patients who had the greatest preservation of retinal structure.[30]

The mechanism for surgically delivering gene therapy to the retina is under much discussion because of the potential trauma subretinal injections may cause, particularly those involving the macula. Some of the phase I/II LCA trials suggested that patients lost visual acuity and retinal thickness after subfoveal injections, potentially due to mechanical trauma to the fovea from inducing a retinal detachment.[11]

Keep in mind that these trials involving subretinal injections are targeting only cells in the region of the surgically induced subretinal bleb, which make up a small percentage of the entire retina (gene therapy clinical trial bleb sizes range from 150 µm to 450 µm).

Zones of retina treated, as well as viral vector dosing, play important roles in the long-term restoration of function. We may yet learn that concomitant neuro-protectant treatments are also going to be useful, if not mandatory, in treating inherited retinal degenerative disease.

Future Trials

AGTC expects to begin enrollment soon of a Phase I/II dose-escalation study for treatment of CNGA3-achromatopsia with AAV (using the same AAV vector and promoter as used in the CNGB3 study).[33] AGTC is also developing an AAV-RPGR vector for X-linked RP for which it plans to sub-

mit an investigational new drug application to the FDA in 2017.[34]

Conclusion

The most thoroughly studied genetic retinal disease at this point remains RPE65-associated LCA2, with multiple Phase I/II trials as well as one Phase III trial completed and one FDA application underway.

Although most phase I/II trials for LCA2 show initial improvement in retinal sensitivity in patients after gene therapy, these improvements were modest even in participants with relatively mild retinal degeneration and failed to protect against ongoing degeneration,[30] suggesting that we still have much room for improvement in the field.

Research into new optimized vectors for therapeutic efficacy and longevity needs to continue. From a clinical standpoint, we still do not fully understand which patients may benefit most from therapy and how therapeutic intervention will alter the natural history of retinal degeneration and progression of vision loss. From a surgical standpoint, more attention is being placed on optimal delivery to minimize mechanical trauma and perioperative inflammation.

Retinal gene therapy has advanced eons in the past 10 years. We will likely see FDA approval in the near future for the first viral-based retinal gene therapy for LCA2. With innovations like optogenetics we can imagine a future where multiple different diseases can be treated with a larger window of opportunity for therapeutic effect. While exciting to the clinical community, these advances will be even more attractive to our patients who, until very recently, have been told at yearly follow-ups, "There is nothing that can be done." We are finally at a point where we can offer realistic hope.

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Is Retina a Window To Bipolar Disorder?

Study evaluates role of retinal photography in psychiatric diagnosis. ***By Melanie R. Naiberg, HBSc, Peter J. Kertes, MD, and Benjamin I. Goldstein, MD, PhD***

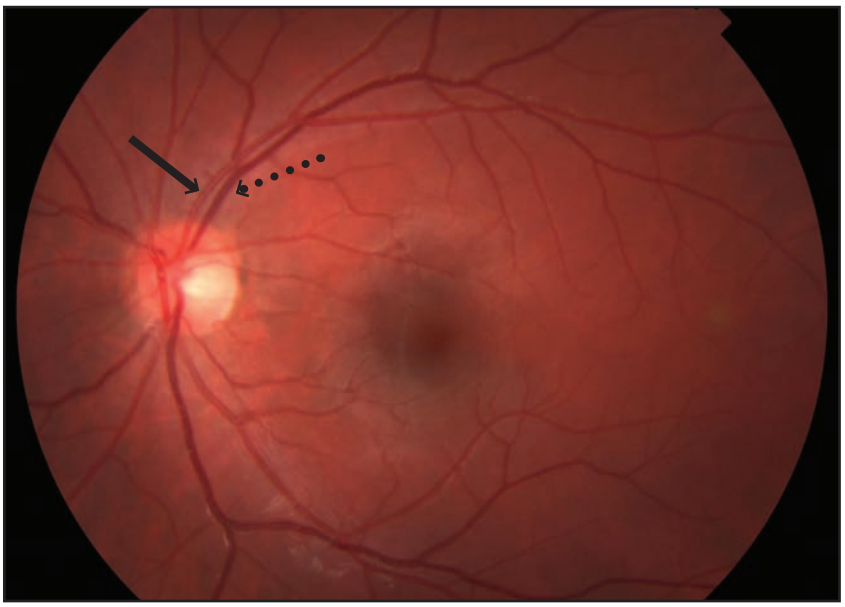
Early detection of psychiatric disorders with retinal imaging would provide a useful non-invasive method for diagnosis and potential treatment early in the disease process. Many studies have looked at, for example, the use of optical coherence tomography (OCT) in Alzheimer's disease.[1]

Another area of research interest is bipolar disorder (BD), an impairing mood disorder characterized by episodes of mania or milder hypomania alternating with depression. Mania consists of grandiosity, decreased need for sleep, flight of ideas, distractibility, increase in goal-directed activity and excessive involvement in activities with high potential for consequences, while depression consists of lack of interest in activities, weight loss or change in appetite, insomnia or hypersomnia, psychomotor agitation or retardation, loss of energy, feelings of worthlessness and diminished ability to concentrate.[2] More severe episodes, symptoms, comorbidities and functional impairment accompany adolescent-onset BD than the adult form.[3,4] Patients with BD also experience a greater prevalence for cardiovascular risk factors (CVRFs) as well as neurocognitive deficits than individuals without BD.[5-8]

Our Canadian research team of psychiatrists and ophthalmologists have looked at retinal photography as a "window to the mind" to see if we can use it as a non-invasive screening tool for psychiatric disorders like BD. Here's what we've learned.

Retina Photography and CVRFs

Early detection of cardiovascular risk prior to the diagnosis of clinical cardiovascular disease (CVD) is crucial. Current techniques for detecting early atherosclerosis proxies include ultrasound to visualize arterial structure (eg, carotid intima media thickness) or function (eg, flow-mediated dilation).[7] However, both techniques are operator-dependent, can be technically challenging and have suboptimal reliability. Additionally, they focus on macrovessels, whereas microvessels may provide a more sensitive assay of early risk of CVD.[9] Given the excessive burden and prematurity of cardiovascular risk in BD, this population has a need for practical, reliable, and cost-effective proxies for measuring atherosclerosis, particularly in adolescents.



Narrow retinal arterioles (solid arrow) and wider retinal venules (dashed arrow) are indicators of cardiovascular risk factors such as diabetes, high blood pressure and dyslipidemia.

Retinal photography provides a noninvasive, inexpensive and reliable method of directly visualizing the retinal microvasculature, which is also a proxy for cerebral microvasculature.[10-12]

Retinal microvessels are arguably the best proxy measures of thcerebrovasculature. First, they can be accurately measured noninvasively. Second, they have several anatomic, embryologic, physiologic and autoregulatory properties similar to cerebral microvasculature.[10,12,13] Third, pathological changes in retinal microcirculation have shown an association with cerebrovascular disease.[11,14]

Current literature indicates that compromised retinal vascular caliber—narrower central retinal arteriolar caliber (CRAE) and wider central retinal venular caliber (CRVE)—is

indicative of various CVRFs, including diabetes, body mass index, obesity, high blood pressure and dyslipidemia.[15-18] Furthermore, investigators have determined that retinal vascular caliber predicts CVD mortality.[19] These associations have also been determined in adolescent populations, where poorer retinal vascular caliber is associated with diabetes, obesity and elevated fasting glucose, body mass index and blood pressure.[20-22]

Cognitive, Psychiatric Disorders

Retinal vascular caliber is also associated with neurocognitive performance among adults. Wider retinal venular caliber is associated with lower IQ and decreased performance on cognitive domains, including verbal comprehension, working memory, processing speed, executive function and motor skills performance.[13] Furthermore, poorer retinal vascular structure has been associated with increased risk for dementia.[23] Studies have also reported that symptoms and diagnosis of depression, as well as schizophrenia, are associated with narrower retinal arteriolar caliber and wider retinal venular caliber.[24-27]

Despite these promising findings, important gaps exist in the literature on retinal vascular photography and how it relates to cardiovascular and neurocognitive aspects. No prior retinal vascular photography studies have focused on BD in any age group, or on neurocognition specifically in adolescents.

Retinal Photography For Integrative Research

The multidisciplinary Canadian study team focused on cardiovascular risk in BD includes ophthalmology, psychiatry, neurology and medical imaging. We are using retinal photography as a novel tool to reliably measure cerebral microvessels.

The goal of this project was to generate through retinal photography novel discoveries about early stages of CVD and neurocognitive impairment in a population highly susceptible to both. This study aimed to examine the association of both CVRFs and neurocognition among adolescents with BD as well as among adolescents who are at high risk for developing BD (that is, they have a parent or a sibling with BD).

We hypothesized that narrower CRAE and wider CRVE would be associated with greater metabolic syndrome components (increased triglycerides, systolic and diastolic blood pressure, glucose and waist circumference, and decreased high-density lipoprotein cholesterol), poorer peripheral endothelial function and poorer frontal-executive neurocognitive task performance.

Thus far the study has enrolled 30 adolescents with BD and 32 healthy controls. A certified ophthalmic assistant performs mydriatic retinal photography to optimize image capture. The study uses anthropometric measures as well as a fasting lipid panel to obtain CVRFs, and bases neurocognitive measures on performance on computer-based tasks using the Cambridge Neuropsychological Tests Automated Battery (CANTAB) software.

Preliminary findings, presented recently at the Society for Biological Psychiatry and Canadian College of Neuropsychopharmacology conferences suggest that poorer retinal vascular caliber is associated with both increased CVRFs and neurocognitive deficits among adolescents with BD, but not in healthy controls. Specifically, higher diastolic blood pressure is associated with lower (worse) arterio-venular ratio (AVR) in the BD group. Similarly, higher (better) endothelial function is associated with higher (better) AVR in the BD group.

With regards to neurocognition, higher (better) AVR is associated with better performance on a task of attention and executive function among BD adolescents. None of these are significant in the health controls.

The theme of early identification and treatment is important in BD, as it is in other medical conditions. Because BD is perhaps the most familial psychiatric condition, examining relatives of people with BD may offer insights that can help parse factors that give rise to or precede BD from those that emerge contemporaneously with symptoms. Adolescent offspring of parents with BD are at an increased risk of developing BD themselves[27-29] and can thus provide insights into underlying cardiovascular risk, independent of the effects of BD.[28,30-32]

If retinal vascular measures are meaningfully associated with BD, CVRFs and neurocognition, one can foresee a future in which retinal vascular photography has practical applications in the diagnosis, monitoring and early treatment of BD. If we succeed in our goal of demonstrating the relevance of retinal vascular measures to early-onset BD, this will

(Continued on page 48)

How Vascular Width Influences Psychological and Cognitive Factors

Wider Retinal Venular Caliber ~ Decreased IQ, decreased cognitive performance, depression, schizophrenia

Narrower Vascular Caliber ~ Depression, schizophrenia

Yes, You Can Negotiate Payer Contracts

You don't always have to take what payers are giving. These steps can protect you from getting shortchanged. ***By Kari Rasmussen***

Sound payer contracts are the foundation of a financially successful practice. Yet, I find that practices don't always scrutinize this important facet of their financial infrastructure with the tenacity it deserves. I often learn of practices that have signed new or revised contracts without so much as reviewing the terms. Here I outline some best practices to consider when evaluating payer contracts.

Negotiate the Terms

Yes, you should try to negotiate the terms. It has been my experience that while most national payers have a take-it-or-leave-it approach, this is not always the case. I've found that regional payers tend to be the most amenable to reconsidering terms for reimbursement.

Regardless of whether the payer is willing to negotiate, a practice has an imperative to scrutinize a payer's contract to determine if the economics make sense. You may be amenable to lower payments if you are trying to garner market share. Conversely, you may decide against taking on a contract if your schedules are already full.

Insurance carriers use their own language, and that could cause you to agree to terms that would be unacceptable if the language were more transparent. Here's an example: "Payer will reimburse Provider for Covered Services at X% of the then prevailing Fee Schedule or X% of the Provider's billed charges, whichever is less."

It sounds very official, but when you distill the language, it does not protect the practice should the payer adjust its prevailing fees. This clause also puts the practice in a position of having to ensure its charge master surpasses the payer's fee schedules.

Know the Payment And Share It

When payers present our practice with a new or revised contract, I send them a list of our most common CPT Codes and ask for their allowed amounts for each plan they are offering to us. This allows us to evaluate whether the terms are agreeable, and we can use this as a basis to negotiate the rate. A critical step in this evaluation process is to make sure that the practice is made whole on expensive biologic agents.

Also, some practices do not make the reimbursement terms available to the key staff members who need that information to do their jobs. We've created a master spreadsheet that outlines the reimbursement rates for our most common CPT codes and we give copies to our staff. This allows them to effectively, and knowledgeably, provide financial counseling, post payments and calculate cost projections for our patients. Without this information, your staff may be writing off charges to your detriment.

Watch the Time-lines

Another piece of payer contracts I look at carefully are the time-lines for claims payment. We insist on payment within 30 days of the payer receiving our claim. We try to include these terms whether the payer is primary or secondary.

Most contracts outline terms for when the practice must submit claims. Most of our contracts require we submit claims within 90 days of the date of service or they will deny them outright. Share these terms with physicians and other providers in your practice who may struggle with timely submission of their charges.

Special Considerations For Medicare Advantage Plans

Medicare Advantage (MA) plans can ask you to accept less than the Medicare allowed amount, but this becomes a contractual decision between you and the payer. Keep in mind, MA plans must cover all services that traditional Medicare covers. The rule of thumb is that MA plans may not cover lesser benefits.

Some payers require providers use specialty pharmacies or step therapy before they'll approve the biologic the doctor prescribes. The contract or provider manuals should outline these terms. Adhering to them will ensure that you are not carelessly using commercially purchased biologics that might not be reimbursed.

Lastly, know what your contract terms are regarding your obligations to provide access to records and audits. Payer audits are on the rise, and knowing your rights can be instrumental in responding to them.

Following these steps can make sure you are effectively scrutinizing your payer contracts so you end up with terms are in the best interest of your practice.

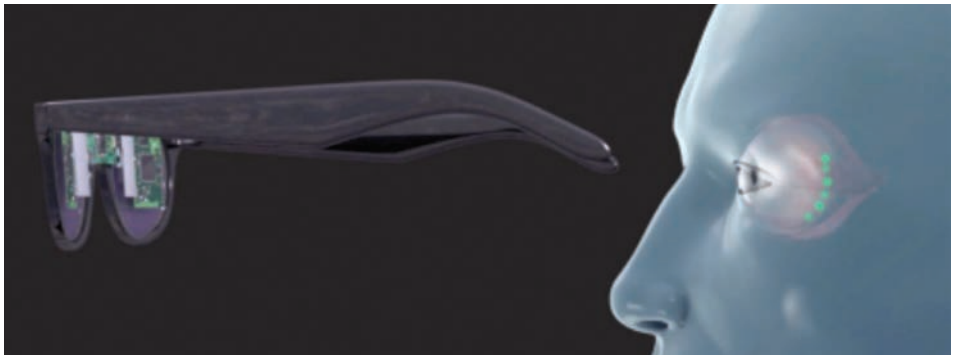
Mr. Laurita is chief operating officer at Retina Associates of Cleveland. Ms. Rasmussen is administrator at Rocky Mountain Retina Consultants, a six-office practice in four western states with the main office in Salt Lake City.

By Richard Mark Kirkner »

A Marriage of Prosthetics, Optogenetics

An investigational neuroprosthetic device pairs with gene therapy to work around damaged photoreceptors.

At the TedMed conference in 2011, Sheila Nirenberg, PhD, a professor of physiology and biophysics at Weill Medical College of Cornell University in New York, talked about her work on a neuroprosthetic device that combines a camera with proprietary software and gene therapies delivered via adeno-associated virus to the back of the eye as a workaround of dysfunctional photoreceptors to convey images to the brain.



A conceptual drawing of the Bionic Sight neuroprosthetic device that is built into a pair of glasses or goggles and relays signals to retinal ganglion cells injected with a transgene to restore vision in people with damaged photoreceptors.

That work led her to start the company Bionic Sight, which formed a strategic collaboration last month with Applied Genetic Technologies Corp. (AGTC), a clinical stage biotechnology company with a pipeline of six gene therapies in ophthalmology. The idea is to use AGTC's knowledge of gene therapy and Bionic Sight's neuroprosthetic device to stimulate the remaining healthy cells in the retina with the retina's neural code to restore normal neural signaling in patients with visual deficits or blindness due to retinal disease.

No Surgery Required

A key part of what makes this treatment different from prosthetic treatments like the Argus II epiretinal prosthesis (Second Sight Medical Products) and others in development is that it does not require any surgery. This is because it doesn't use electrodes to stimulate cells, but instead uses optogenetics, which only requires an injection into the patient's eye. A vector carrying the gene for an optogenetic protein is injected into the eye and taken up by the ganglion cells, and the optogenetic protein can then be used to drive the cells to send signals to the brain.

The neuroprosthetic treatment consists of two parts: the device and the optogenetic protein. "The device takes images in and converts them into the retina's code," Dr. Nirenberg says. "It does this by performing a transformation on them that turns them into the same patterns of action potentials that the normal retina would produce; that is, it converts them into the language of the brain."

The signals are then sent to the ganglion cells, which use the optogenetic protein to send the signals forward, so they reach the brain.

Bypassing the Problem

The concept is not to target damaged photoreceptors. "Instead it is to take patients with retinal degenerative diseases, who have very damaged photoreceptors, and bypass the problem by going right to the ganglion cells," Dr. Nirenberg says. The ganglion cells then send the code directly on to the brain. AGTC would develop adeno-associated virus-based gene therapies to deliver those optogenetic vectors into the ganglion cells.

This approach is unique because typical gene-replacement therapy requires accurate identification of the damaged gene and then replacing it with new genetic material. "With this approach, it doesn't matter which gene caused the problem; as long as the patient still has functioning ganglion cells, we'll jump right over the damaged photoreceptors and go right to the final-stage output cells," Dr. Nirenberg says. However, this approach won't work in glaucoma because the ganglion cells are damaged.

The research with the device is still in the preclinical stage, but Dr. Nirenberg says the team of investigators is finishing the remaining safety studies and is in the process of compiling a pre-investigational new drug application for the Food and Drug Administration.

Edited by Emmett T. Cunningham Jr., MD, PhD»

Can a Gene Vector Work for Long Term?

Lentiviral vector expresses protein for up to four years in trial.

A viral vector for gene therapy to treat neovascular age-related macular degeneration has demonstrated long-term safety of gene transfer and therapeutic protein production for up to four years in the eyes of affected patients. Recent Phase I trial results of the vector may lead to packaging of a protein that binds vascular endothelial growth factor.

The trial evaluated RetinoStat (Oxford BioMedica, Oxford, UK), a lentiviral equine infectious anemia virus (EIAV) vector that expressed endostatin and angiostatin in 21 patients with neovascular AMD. Results were published in the January issue of *Human Gene Therapy*.[1]

Patients were enrolled in three different dose-ranging cohorts—2.4×10^4 transduction units (TU); 2.4×10^5 TU; and 8×10^5 TU, each with three patients—and one cohort of 12 receiving the highest dose of 8×10^5 TU. The study duration was 48 weeks and patients were encouraged to enroll in a 15-year long-term follow-up study.

All of the doses were well tolerated, although one patient developed a macular hole that was treated and resolved. The study documented a dose-related increase in levels of endostatin and angiostatin in the aqueous humor, which peaked between weeks 12 and 24 in the highest-dose group.

Fluorescein angiography showed a reduction in leakage in most patients, but only one subject showed convincing evidence of anti-permeability. Lesion size, however, showed no significant change in the maximum therapy group.

Here, principal investigator Peter A. Campochiaro, MD, Eccles Professor of Ophthalmology and Neuroscience at Johns Hopkins Wilmer Eye Institute, Baltimore, discusses the Lentiviral Vector Gene Transfer of Endostatin/Angiostatin for Macular Degeneration (GEM) study.

Summary of GEM findings in his own words:

RetinoStat is an equine infectious anemia lentiviral vector that enters cells and produces endostatin and angiostatin, both of which are proteins that have strongly suppressed abnormal blood vessel growth and leakage in mouse models of wet AMD. Several studies have demonstrated that subretinal injection of adeno-associated viral (AAV) vectors is safe and well-tolerated.

The purpose of the GEM trial was to determine if subretinal injection of a lentiviral vector is safe and well-tolerated in patients with advanced wet AMD.

Lentiviral vectors differ from AAV vectors in that they incorporate into the genome, meaning that they insert into a patient's DNA, which is the most stable situation for long-term expression. The study demonstrated that subretinal injection of an EIAV vector caused no identifiable toxicity and thus was safe.

Regarding secondary trial endpoints:

The major secondary endpoint was to determine if there was good production of endostatin and angiostatin. Levels of endostatin and angiostatin in the aqueous humor were measured at several time points after subretinal injection. We did this by inserting a needle into the front of the eye and removing a small amount of fluid used to measure levels of the proteins.

High levels of endostatin and angiostatin were present in the aqueous humor obtained from almost all patients, and the levels did not decrease over time. Many patients had measurements years after the subretinal injection of RetinoStat and the levels of endostatin and angiostatin were still high with no decrease from earlier levels.

Other secondary endpoints were the measurement of the amount of fluid in the macula by optical coherence tomography and change in visual acuity from baseline. There was no significant reduction in fluid or improvement of visual acuity after injection of RetinoStat—but most patients had very advanced wet AMD with scarring in the macula resulting in permanent loss of vision that could not be improved.

The mechanism of action of RetinoStat in his own words:

When RetinoStat is injected under the retina, it enters retinal pigment epithelium cells and photoreceptors, inserts into the DNA of those cells and begins producing endostatin and angiostatin. We know that this occurred because after the injection of RetinoStat, patients had high levels of endostatin and angiostatin in their eyes

and before the injection they did not.

In mice, endostatin and angiostatin cause reduction in abnormal blood vessels and leakage of fluid from abnormal blood vessels. Our study suggests that endostatin and angiostatin may have less effect on abnormal blood vessels in humans than they do in mice.

What investigators learned about the potential for changes in visual acuity:

There were a few patients who had improvement in vision and reduced fluid after the injection of RetinoStat, but the majority did not.

How findings may inform future trials:

This study indicates that EIAV is an extremely good platform for gene therapy because it provides long-lasting production of proteins.

Next steps in RetinoStat development:

A potential next step is to package a protein that binds vascular endothelial growth factor (VEGF) in EIAV. We already know that injections of proteins that bind VEGF provide benefit in patients with wet AMD. Long-term production of such proteins in the eye after injection of a lentiviral vector could provide a long-term benefit. RS

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(Continued from page 44)

provide the rationale for further development of this pragmatic method of early identification and monitoring of BD. In particular, we intend to integrate vascular-related neuroimaging phenotypes and longitudinal repeated measures. This study will provide an excellent platform for future, larger-scale, longitudinal studies. Overall, this study comprises an important step toward better understanding adolescent BD and its relation to increased cardiovascular risk and neurocognitive deficits. RS

Dr. Mandelcorn is an assistant professor of ophthalmology at the University of Toronto, where Ms. Naiberg is also a PhD candidate, Dr. Kertes a vitreoretinal surgeon and Dr. Goldstein an associate professor of psychiatry and pharmacology.

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Professional Opportunities

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Bassett Healthcare Network, a progressive health care network in central New York and major teaching affiliate of Columbia University, is seeking a Medical Retina Specialist to join a busy ophthalmology department based in the lovely village of Cooperstown.

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EEO EMPLOYER

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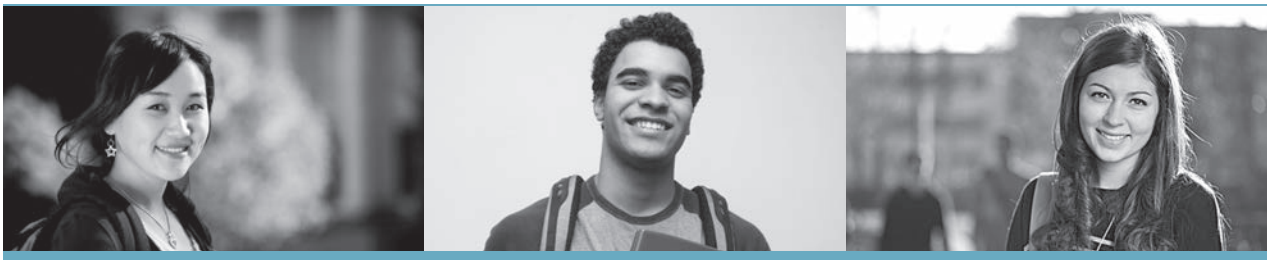

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EYLEA®
(aflibercept) Injection
For Intravitreal Injection

BRIEF SUMMARY OF FULL PRESCRIBING INFORMATION

FOR COMPLETE DETAILS, SEE FULL PRESCRIBING INFORMATION.

1 INDICATIONS AND USAGE

EYLEA® (aflibercept) Injection is indicated for the treatment of patients with Neovascular (Wet) Age-Related Macular Degeneration (AMD), Macular Edema following Retinal Vein Occlusion (RVO), Diabetic Macular Edema (DME), and Diabetic Retinopathy (DR) in Patients with DME.

2 DOSAGE AND ADMINISTRATION

2.1 Important Injection Instructions. For ophthalmic intravitreal injection. EYLEA must only be administered by a qualified physician.

2.2 Neovascular (Wet) Age-Related Macular Degeneration (AMD). The recommended dose for EYLEA is 2 mg (0.05 mL or 50 microliters) administered by intravitreal injection every 4 weeks (monthly) for the first 12 weeks (3 months), followed by 2 mg (0.05 mL) via intravitreal injection once every 8 weeks (2 months). Although EYLEA may be dosed as frequently as 2 mg every 4 weeks (monthly), additional efficacy was not demonstrated in most patients when EYLEA was dosed every 4 weeks compared to every 8 weeks. Some patients may need every 4 week (monthly) dosing after the first 12 weeks (3 months).

2.3 Macular Edema Following Retinal Vein Occlusion (RVO). The recommended dose for EYLEA is (0.05 mL or 50 microliters) administered by intravitreal injection once every 4 weeks (monthly).

2.4 Diabetic Macular Edema (DME). The recommended dose for EYLEA is (0.05 mL or 50 microliters) administered by intravitreal injection every 4 weeks (monthly) for the first 5 injections followed by 2 mg (0.05 mL) via intravitreal injection once every 8 weeks (2 months). Although EYLEA may be dosed as frequently as 2 mg every 4 weeks (monthly), additional efficacy was not demonstrated in most patients when EYLEA was dosed every 4 weeks compared to every 8 weeks. Some patients may need every 4 week (monthly) dosing after the first 20 weeks (5 months).

2.5 Diabetic Retinopathy (DR) in Patients with DME. The recommended dose for EYLEA is 2 mg (0.05 mL or 50 microliters) administered by intravitreal injection every 4 weeks (monthly) for the first 5 injections, followed by 2 mg (0.05 mL) via intravitreal injection once every 8 weeks (2 months). Although EYLEA may be dosed as frequently as 2 mg every 4 weeks (monthly), additional efficacy was not demonstrated in most patients when EYLEA was dosed every 4 weeks compared to every 8 weeks. Some patients may need every 4 week (monthly) dosing after the first 20 weeks (5 months).

2.6 Preparation for Administration. EYLEA should be inspected visually prior to administration. If particulates, cloudiness, or discoloration are visible, the vial must not be used. Using aseptic technique, the intravitreal injection should be performed with a 30-gauge x ½-inch injection needle. For complete preparation for administration instructions, see full prescribing information.

2.7 Injection Procedure. The intravitreal injection procedure should be carried out under controlled aseptic conditions, which include surgical hand disinfection and the use of sterile gloves, a sterile drape, and a sterile eyelid speculum (or equivalent). Adequate anesthesia and a topical broad–spectrum microbicide should be given prior to the injection.

Immediately following the intravitreal injection, patients should be monitored for elevation in intraocular pressure. Appropriate monitoring may consist of a check for perfusion of the optic nerve head or tonometry. If required, a sterile paracentesis needle should be available.

Following intravitreal injection, patients should be instructed to report any symptoms suggestive of endophthalmitis or retinal detachment (e.g., eye pain, redness of the eye, photophobia, blurring of vision) without delay (*see Patient Counseling Information*).

Each vial should only be used for the treatment of a single eye. If the contralateral eye requires treatment, a new vial should be used and the sterile field, syringe, gloves, drapes, eyelid speculum, filter, and injection needles should be changed before EYLEA is administered to the other eye.

After injection, any unused product must be discarded.

3 DOSAGE FORMS AND STRENGTHS

Single-use, glass vial designed to provide 0.05 mL of 40 mg/mL solution (2 mg) for intravitreal injection.

4 CONTRAINDICATIONS

EYLEA is contraindicated in patients with

- Ocular or periocular infections
- Active intraocular inflammation
- Known hypersensitivity to aflibercept or any of the excipients in EYLEA.

Hypersensitivity reactions may manifest as severe intraocular inflammation.

5 WARNINGS AND PRECAUTIONS

5.1 Endophthalmitis and Retinal Detachments. Intravitreal injections, including those with EYLEA, have been associated with endophthalmitis and retinal detachments (*see Adverse Reactions*). Proper aseptic injection technique must always be used when administering EYLEA. Patients should be instructed to report any symptoms suggestive of endophthalmitis or retinal detachment without delay and should be managed appropriately (*see Dosage and Administration and Patient Counseling Information*).

5.2 Increase in Intraocular Pressure. Acute increases in intraocular pressure have been seen within 60 minutes of intravitreal injection, including with EYLEA (*see Adverse Reactions*). Sustained increases in intraocular pressure have also been reported after repeated intravitreal dosing with vascular edothelial growth factor (VEGF) inhibitors. Intraocular pressure and the perfusion of the optic nerve head should be monitored and managed appropriately (*see Dosage and Administration*).

5.3 Thromboembolic Events. There is a potential risk of arterial thromboembolic events (ATEs) following intravitreal use of VEGF inhibitors, including EYLEA. ATEs are defined as nonfatal stroke, nonfatal myocardial infarction, or vascular death (including deaths of unknown cause). The incidence of reported thromboembolic events in wet AMD studies during the first year was 1.8% (32 out of 1824) in the combined group of patients treated with EYLEA. The incidence in the DME studies from baseline to week 52 was 3.3% (19 out of 578) in the combined group of patients treated with EYLEA compared with 2.8% (8 out of 287) in the control group; from baseline to week 100, the incidence was 6.4% (37 out of 578) in the combined group of patients treated with EYLEA compared with 4.2% (12 out of 287) in the control group. There were no reported thromboembolic events in the patients treated with EYLEA in the first six months of the RVO studies.

6 ADVERSE REACTIONS

The following adverse reactions are discussed in greater detail in the *Warnings and Precautions* section of the labeling:

- Endophthalmitis and retinal detachments
- Increased intraocular pressure
- Thromboembolic events

6.1 Clinical Trials Experience. Because clinical trials are conducted under widely varying conditions, adverse reaction rates observed in the clinical trials of a drug cannot be directly compared to rates in other clinical trials of the same or another drug and may not reflect the rates observed in practice.

A total of 2711 patients treated with EYLEA constituted the safety population in seven phase 3 studies. Among those, 2110 patients were treated with the recommended dose of 2 mg. Serious adverse reactions related to the injection procedure have occurred in <0.1% of intravitreal injections with EYLEA including endophthalmitis and retinal detachment. The most common adverse reactions (≥5%) reported in patients receiving EYLEA were conjunctival hemorrhage, eye pain, cataract, vitreous floaters, intraocular pressure increased, and vitreous detachment.

Neovascular (Wet) Age-Related Macular Degeneration (AMD). The data described below reflect exposure to EYLEA in 1824 patients with wet AMD, including 1223 patients treated with the 2-mg dose, in 2 double-masked, active-controlled clinical studies (VIEW1 and VIEW2) for 12 months.

Table 1: Most Common Adverse Reactions (≥1%) in Wet AMD Studies

Adverse Reactions	EYLEA (N=1824)	Active Control (ranibizumab) (N=595)
Conjunctival hemorrhage	25%	28%
Eye pain	9%	9%
Cataract	7%	7%
Vitreous detachment	6%	6%
Vitreous floaters	6%	7%
Intraocular pressure increased	5%	7%
Ocular hyperemia	4%	8%
Corneal epithelium defect	4%	5%
Detachment of the retinal pigment epithelium	3%	3%
Injection site pain	3%	3%
Foreign body sensation in eyes	3%	4%
Lacrimation increased	3%	1%
Vision blurred	2%	2%
Intraocular inflammation	2%	3%
Retinal pigment epithelium tear	2%	1%
Injection site hemorrhage	1%	2%
Eyelid edema	1%	2%
Corneal edema	1%	1%

Less common serious adverse reactions reported in <1% of the patients treated with EYLEA were hypersensitivity, retinal detachment, retinal tear, and endophthalmitis.

Macular Edema Following Retinal Vein Occlusion (RVO). The data described below reflect 6 months exposure to EYLEA with a monthly 2 mg dose in 218 patients following CRVO in 2 clinical studies (COPERNICUS and GALILEO) and 91 patients following BRVO in one clinical study (VIBRANT).

Table 2: Most Common Adverse Reactions (≥1%) in RVO Studies

Adverse Reactions	CRVO		BRVO	
	EYLEA (N=218)	Control (N=142)	EYLEA (N=91)	Control (N=92)
Eye pain	13%	5%	4%	5%
Conjunctival hemorrhage	12%	11%	20%	4%
Intraocular pressure increased	8%	6%	2%	0%
Corneal epithelium defect	5%	4%	2%	0%
Vitreous floaters	5%	1%	1%	0%
Ocular hyperemia	5%	3%	2%	2%
Foreign body sensation in eyes	3%	5%	3%	0%
Vitreous detachment	3%	4%	2%	0%
Lacrimation increased	3%	4%	3%	0%
Injection site pain	3%	1%	1%	0%
Vision blurred	1%	<1%	1%	1%
Intraocular inflammation	1%	1%	0%	0%
Cataract	<1%	1%	5%	0%
Eyelid edema	<1%	1%	1%	0%

Less common adverse reactions reported in <1% of the patients treated with EYLEA in the CRVO studies were corneal edema, retinal tear, hypersensitivity, and endophthalmitis.

Diabetic Macular Edema (DME). The data described below reflect exposure to EYLEA in 578 patients with DME treated with the 2-mg dose in 2 double-masked, controlled clinical studies (VIVID and VISTA) from baseline to week 52 and from baseline to week 100.

Table 3: Most Common Adverse Reactions (≥1%) in DME Studies

Adverse Reactions	Baseline to Week 52		Baseline to Week 100	
	EYLEA (N=578)	Control (N=287)	EYLEA (N=578)	Control (N=287)
Conjunctival hemorrhage	28%	17%	31%	21%
Eye pain	9%	6%	11%	9%
Cataract	8%	9%	19%	17%
Vitreous floaters	6%	3%	8%	6%
Corneal epithelium defect	5%	3%	7%	5%
Intraocular pressure increased	5%	3%	9%	5%
Ocular hyperemia	5%	6%	5%	6%
Vitreous detachment	3%	3%	8%	6%
Foreign body sensation in eyes	3%	3%	3%	3%
Lacrimation increased	3%	2%	4%	2%
Vision blurred	2%	2%	3%	4%
Intraocular inflammation	2%	<1%	3%	1%
Injection site pain	2%	<1%	2%	<1%
Eyelid edema	<1%	1%	2%	1%

Less common adverse reactions reported in <1% of the patients treated with EYLEA were hypersensitivity, retinal detachment, retinal tear, corneal edema, and injection site hemorrhage.

6.2 Immunogenicity. As with all therapeutic proteins, there is a potential for an immune response in patients treated with EYLEA. The immunogenicity of EYLEA was evaluated in serum samples. The immunogenicity data reflect the percentage of patients whose test results were considered positive for antibodies to EYLEA in immunoassays. The detection of an immune response is highly dependent on the sensitivity and specificity of the assays used, sample handling, timing of sample collection, concomitant medications, and underlying disease. For these reasons, comparison of the incidence of antibodies to EYLEA with the incidence of antibodies to other products may be misleading.

In the wet AMD, RVO, and DME studies, the pre-treatment incidence of immunoreactivity to EYLEA was approximately 1% to 3% across treatment groups. After dosing with EYLEA for 24-100 weeks, antibodies to EYLEA were detected in a similar percentage range of patients. There were no differences in efficacy or safety between patients with or without immunoreactivity.

6.3 Postmarketing Experience. The following adverse reactions have been identified during postapproval use of EYLEA. Because these reactions are reported voluntarily from a population of uncertain size, it is not always possible to reliably estimate their frequency or establish a causal relationship to drug exposure.

- Hypersensitivity including rash, pruritus, and urticaria as well as isolated cases of severe anaphylactic/anaphylactoid reactions.

8 USE IN SPECIFIC POPULATIONS

8.1 Pregnancy. Pregnancy Category C. Aflibercept produced embryo-fetal toxicity when administered every three days during organogenesis to pregnant rabbits at intravenous doses ≥3 mg per kg, or every six days at subcutaneous doses ≥0.1 mg per kg. Adverse embryo-fetal effects included increased incidences of postimplantation loss and fetal malformations, including anasarca, umbilical hernia, diaphragmatic hernia, gastroschisis, cleft palate, ectrodactyly, intestinal atresia, spina bifida, encephalomeningocele, heart and major vessel defects, and skeletal malformations (fused vertebrae, sternebrae, and ribs; supernumerary vertebral arches and ribs; and incomplete ossification). The maternal No Observed Adverse Effect Level (NOAEL) in these studies was 3 mg per kg. Aflibercept produced fetal malformations at all doses assessed in rabbits and the fetal NOAEL was less than 0.1 mg per kg. Administration of the lowest dose assessed in rabbits (0.1 mg per kg) resulted in systemic exposure (AUC) that was approximately 10 times the systemic exposure observed in humans after an intravitreal dose of 2 mg.

There are no adequate and well-controlled studies in pregnant women. EYLEA should be used during pregnancy only if the potential benefit justifies the potential risk to the fetus. Females of reproductive potential should use effective contraception prior to the initial dose, during treatment, and for at least 3 months after the last intravitreal injection of EYLEA.

8.3 Nursing Mothers. It is unknown whether aflibercept is excreted in human milk. Because many drugs are excreted in human milk, a risk to the breastfed child cannot be excluded. EYLEA is not recommended during breastfeeding. A decision must be made whether to discontinue nursing or to discontinue treatment with EYLEA, taking into account the importance of the drug to the mother.

8.4 Pediatric Use. The safety and effectiveness of EYLEA in pediatric patients have not been established.

8.5 Geriatric Use. In the clinical studies, approximately 76% (2049/2701) of patients randomized to treatment with EYLEA were ≥65 years of age and approximately 46% (1250/2701) were ≥75 years of age. No significant differences in efficacy or safety were seen with increasing age in these studies.

17 PATIENT COUNSELING INFORMATION

In the days following EYLEA administration, patients are at risk of developing endophthalmitis or retinal detachment. If the eye becomes red, sensitive to light, painful, or develops a change in vision, advise patients to seek immediate care from an ophthalmologist (*see Warnings and Precautions*). Patients may experience temporary visual disturbances after an intravitreal injection with EYLEA and the associated eye examinations (*see Adverse Reactions*). Advise patients not to drive or use machinery until visual function has recovered sufficiently.

REGENERON

Manufactured by:
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June 2016

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DORC's history in innovation is changing the surgical landscape.

- **TDC: two-dimensional cutter**
  - Up to 16,000 cpm*
  - Port open design enhances surgeon efficiency & predictability
- **Dual capabilities**
  - Phaco – MICS 1.8 capable
  - Vitrectomy – 20g, 23g, 25g & 27g options
- **VTi fluidics**
  - Linear Displacement Pump provides surgeons with enhanced stability & control
  - Vacuum or Flow options on the fly
- **LED illumination**
  - 10,000 hours of consistent light output

DUTCH OPHTHALMIC USA

D.O.R.C. International

Dutch Ophthalmic Research Center

* The TDC vitrectome cuts at 8,000 cpm and is designed to cut on the return of each stroke, doubling the cut rate.

Discover Strength in efficacy

As demonstrated in phase 3 clinical trials in patients with Wet AMD, Macular Edema following RVO, DME, and DR in patients with DME

Choose EYLEA® (aflibercept) Injection from the start

Learn about EYLEA at EYLEA.us/ro

INDICATIONS AND IMPORTANT SAFETY INFORMATION

INDICATIONS

- EYLEA® (aflibercept) Injection is indicated for the treatment of patients with Neovascular (Wet) Age-related Macular Degeneration (AMD), Macular Edema following Retinal Vein Occlusion (RVO), Diabetic Macular Edema (DME), and Diabetic Retinopathy (DR) in Patients with DME.

CONTRAINDICATIONS

- EYLEA® (aflibercept) Injection is contraindicated in patients with ocular or periocular infections, active intraocular inflammation, or known hypersensitivity to aflibercept or to any of the excipients in EYLEA.

WARNINGS AND PRECAUTIONS

- Intravitreal injections, including those with EYLEA, have been associated with endophthalmitis and retinal detachments. Proper aseptic injection technique must always be used when administering EYLEA. Patients should be instructed to report any symptoms suggestive of endophthalmitis or retinal detachment without delay and should be managed appropriately. Intraocular inflammation has been reported with the use of EYLEA.
- Acute increases in intraocular pressure have been seen within 60 minutes of intravitreal injection, including with EYLEA. Sustained increases in intraocular pressure have also been reported after repeated intravitreal dosing with VEGF inhibitors. Intraocular pressure and the perfusion of the optic nerve head should be monitored and managed appropriately.
- There is a potential risk of arterial thromboembolic events (ATEs) following intravitreal use of VEGF inhibitors, including EYLEA. ATEs are defined as nonfatal stroke, nonfatal myocardial infarction, or vascular death (including deaths of unknown cause). The incidence of reported thromboembolic events in wet AMD studies during the first year was 1.8% (32 out of 1824) in the combined group of patients treated with EYLEA. The incidence in the DME studies from baseline to week 52 was 3.3% (19 out of 578) in the combined group of patients treated with EYLEA compared with 2.8% (8 out of 287) in the control group; from baseline to week 100, the incidence was 6.4% (37 out of 578) in the combined group of patients treated with EYLEA compared with 4.2% (12 out of 287) in the control group. There were no reported thromboembolic events in the patients treated with EYLEA in the first six months of the RVO studies.

ADVERSE REACTIONS

- Serious adverse reactions related to the injection procedure have occurred in <0.1% of intravitreal injections with EYLEA including endophthalmitis and retinal detachment.
- The most common adverse reactions (≥5%) reported in patients receiving EYLEA were conjunctival hemorrhage, eye pain, cataract, vitreous floaters, intraocular pressure increased, and vitreous detachment.

Please see brief summary of full Prescribing Information on the following page.

EYLEA®
(aflibercept) Injection
For Intravitreal Injection

TARGETED SCIENCE

REGENERON

06/2016
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