Gene Therapy for Retinal Disease

David M Brown MD
Retinal Consultants of Houston

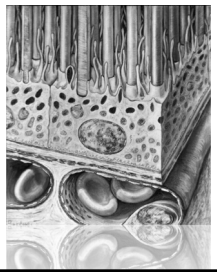
Relevant Disclosures

- Consultant / Grant Support: Allergan, Apellis, Genentech / Roche, Novartis, Regeneron, REGENXBIO, Adverum, Clearside Biomedical, Opthea, Samsung, Santen, Bayer, Senju, Zeiss, Heidelberg, OHR, BioTime, Gemini, Chengdu Kanghong Biotechnology, Optos, Kodiak Sciences, Johnson & Johnson
- Co-patent holder on OPTOS de-warping algorithms

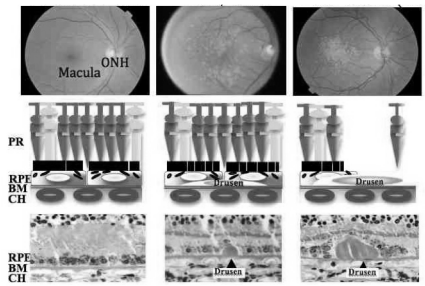
DMB had full control of the presentation

Target of Stem Cell Therapy: RPE

- Sustains photoreceptors
- Highly differentiated monolayer
- Polarized but non-synaptic
- Surgically accessible
- Direct visualization with multimodal (FA/OCT) imaging in the clinic

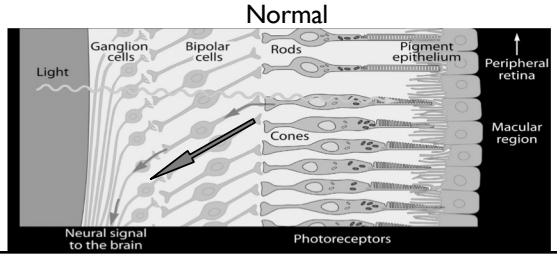


RPE Dysfunction Leads to Photoreceptor Death



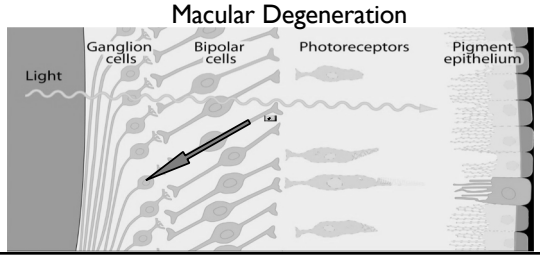
Visual Physiology: Normal vs Macular Degeneration

Normal

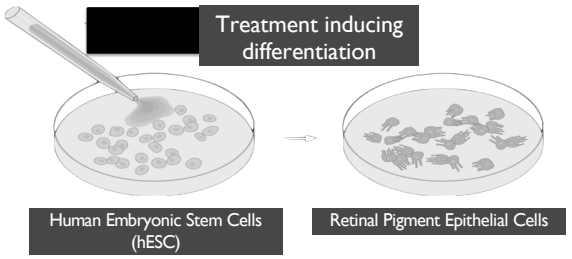


Visual Physiology: Normal vs Macular Degeneration

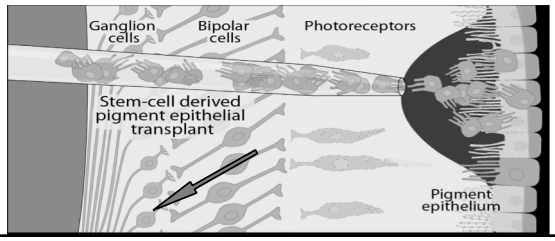
Macular Degeneration



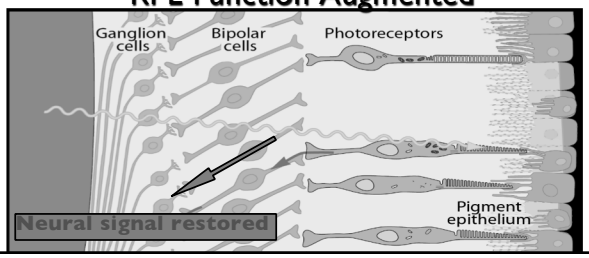
Stem Cell Therapy



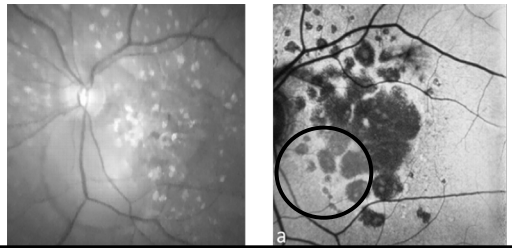
Thesis of RPE Transplantation: Restore Anatomy and Function

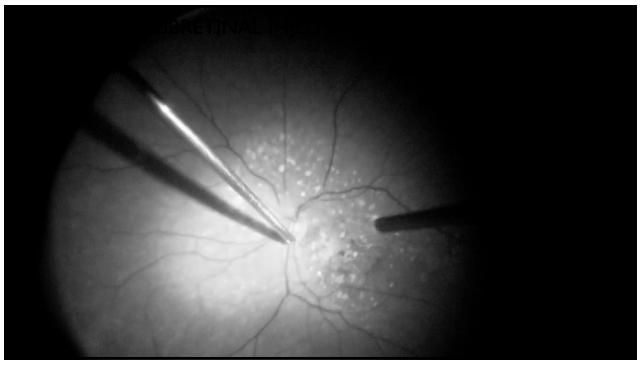


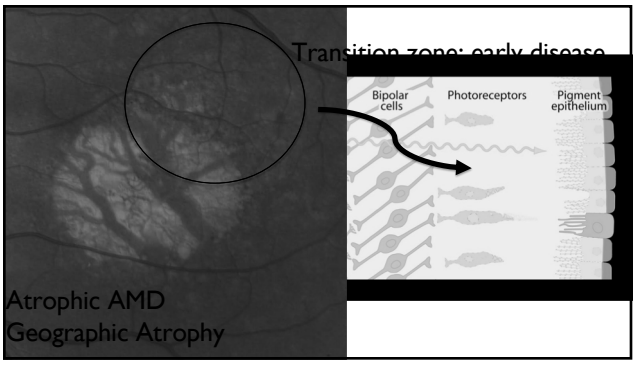
Thesis of RPE Transplantation: RPE Function Augmented

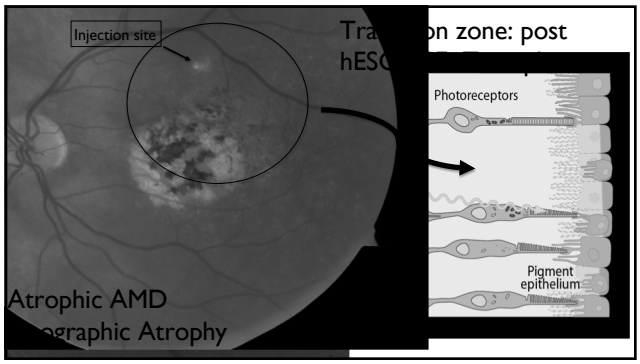


Transplant site selection:
Transition zone between areas of atrophy and normal RPE

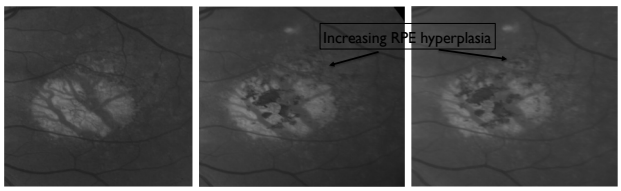




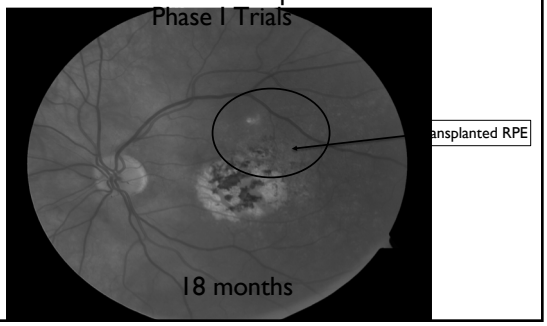




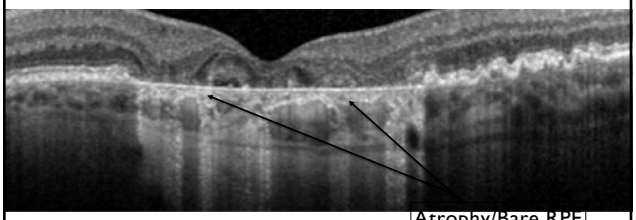
RPE Stem Cell Transplantation
Phase I Trials



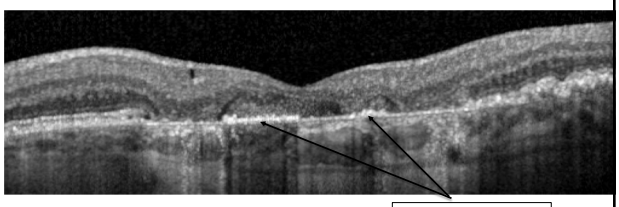
RPE Stem Cell Transplantation
Phase I Trials

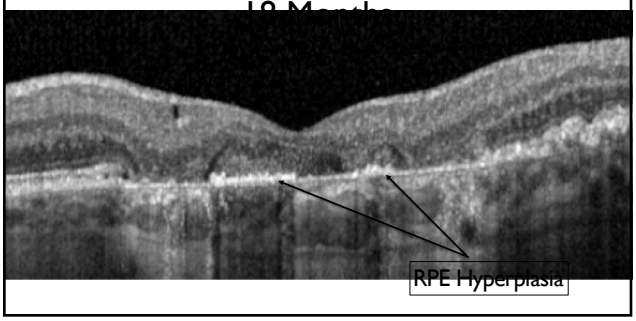


Baseline

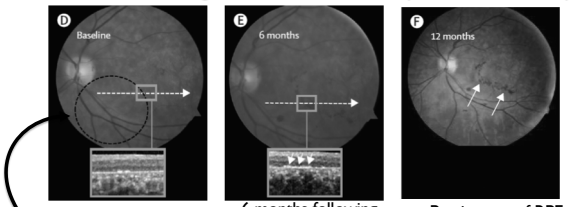


6 Months





Phase 1/2 Trials: Evidence of Engraftment 13/18 (Lancet, 2014)



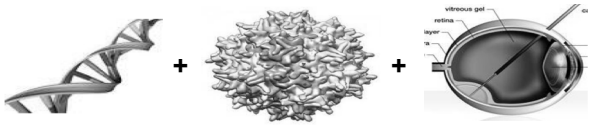
area of subretinal transplantation

6 months following subretinal MA09-hRPE injection

Persistence of RPE 12 months post-transplantation

Why is the retina readily amendable to gene therapy?

GENE + AAV VECTOR + TARGET



Protein Based Therapies eg Anti-VEGF or Complement System Inhibitors

AAV Family Non pathogenic Escape Immunity Tissue Tropism

Immune Privilege Surgical Access

Leber's Congenital Amuarosis

- First described in 1869 by Theodore Leber.
- Rare retinal disease typically inherited in a recessive manner
- Affects 1 in 80,000 individuals.
- Severe loss in vision at birth or in first few months of life.

Leber's Congenital Amuarosis Mutations in RPE65

- Inherited mutations in the RPE65 gene result in either the absence or the presence of a dysfunctional protein.

- Delivery of the normal LCA2 gene into RPE cells should express a normal RPE65 protein that can function in Vitamin A metabolism enabling photoreceptor cells to function.

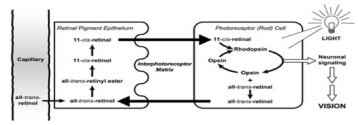
Animal Models for LCA 2 (RPE65)

- Mice have been developed in which the RPE65 gene has been knocked out or inactivated.
- Briard dog which has inherited mutations in RPE65.

Both these animal models have severe loss in vision but retain some of their photoreceptor cells

Successful Gene Therapy for LCA Type 2 Using RPE65 Gene

AAV2 Vector transfecting RPE cells with RPE65 gene with stable expression



Spark Therapeutics

Briard Dog with LCA Treated vs Untreated

Gene Therapy Clinical Trails for Leber's Congenital Amaurosis

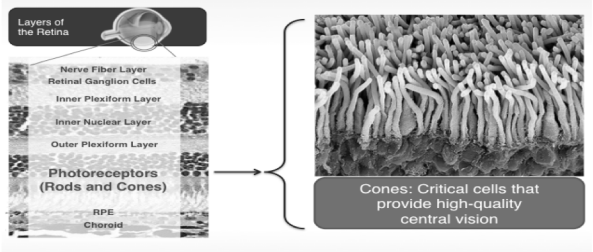
- Three human Phase I clinical trials started in 2008
  - 1 in London and 2 in Phila, PA
  - Others are planned and will follow shortly
- Initial Studies on 9 young adults (19 – 26 yrs old) with LCA2 have been reported
  - All had significant visual loss, but some remaining photoreceptors

Retinal Diseases with Successful Gene Delivery Models in Animals

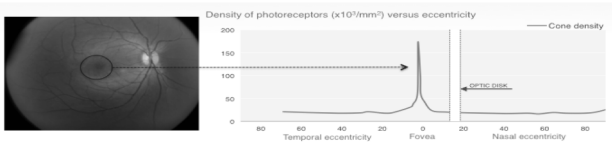
- X-linked Retinoschisis
- Recessive RP
- Cone Dystrophies
- Other forms of LCA
- Stargardt's Macular Degeneration

Continued success of the LCA-RPE65 clinical trials will facilitate new clinical trials in some of these diseases

Cone Photoreceptors: Key Target Cells for Central Retina

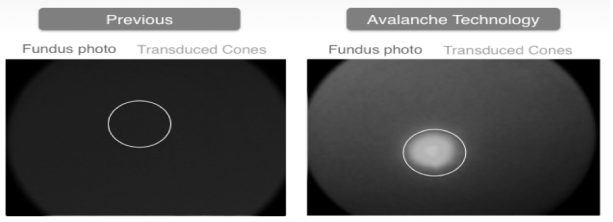


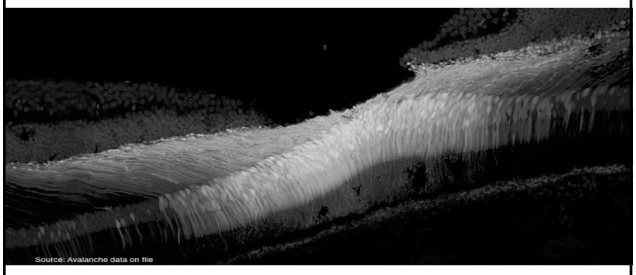
Cone Photoreceptors: High Density in Central Retina



- HD central vision
- Daytime acuity
- Color discrimination

Efficient Cone Delivery in Primates





Biology of Normal Color Vision

Each cone cell makes one of three photopigments:

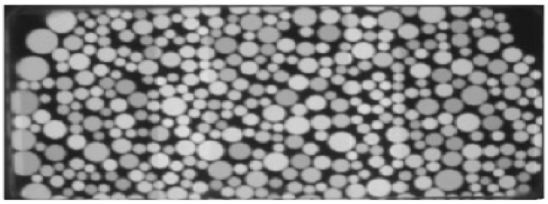
BLUE CONES	GREEN CONES	RED CONES
5%	5-90%	5-90%

Biology of Normal Color Vision

Biology of Abnormal Color Vision

Color Blindness in Squirrel Monkeys

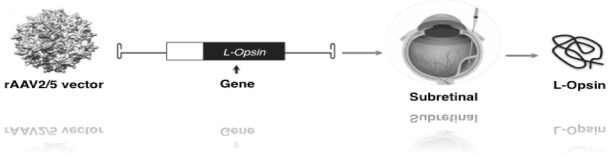
Would the presence of L-Opsin allow the animals to see new colors?



Cambridge Colour Test

Dalton: Color Blind

Gene Therapy with L-Opsin Delivered Subretinally



Dalton: Weeks Post Gene Therapy

Dalton: Several Months Later

Duration Effect > 3 Years from a Single Injection

Long-Term Results of Gene Therapy for Red-Green Color Blindness in Monkeys

K. Mancuso[1A], M. Neitz[1A], W. W. Hauswirth[2], Q. Li[3], T. B. Connor[4], J. A. Kuchenbecker[1], M. C. Mauck[5] and J. Neitz[6]

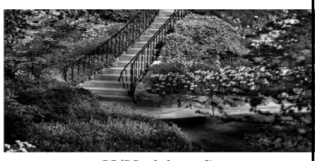
[A]Ophthalmology,[1]University of Washington, Seattle, Washington
[2]Dept of Ophthalmology, Univ of Florida Coll of Medicine, Gainesville, Florida
[3]Ophthalmology, University of Florida, Gainesville, Florida
[4]Vitreoretinal Service, Eye Institute Med Coll WI, Milwaukee, Wisconsin
[5]Ophthalmology, Medical College of Wisconsin, Milwaukee, Wisconsin
[6]Ophthalmology, Univ of Washington, Medical School, Seattle, Washington

Commercial Relationships: K. Mancuso, None; **M. Neitz**, None; **W.W. Hauswirth**, AGTC, Inc. (Alachua, FL), C; **Q. Li**, None; **T.B. Connor**, None; **J.A. Kuchenbecker**, None; **M.C. Mauck**, None; **J. Neitz**, None.

Support: R01EY016861, R03EY014056, R01EY11123, T32EY014537, P30EY01931, P30EY01730, P30EY08571, RR00166, Heeb Fdn., Posner Fdn., Macular Vis. Res. Fdn., Fdn. for Fighting Blindness, Hope for Vis., and RPB

3 Million Americans Have Cataract Surgery Every Year

20/50 vision before



20/20 vision after

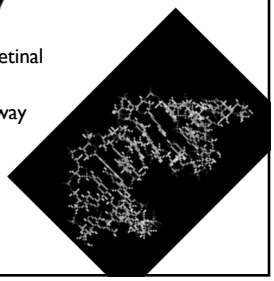
Approx. 10 Million Americans Color Blind

Color vision deficiency before

Normal color vision after

Gene Therapy Summary

- Under development for inherited retinal diseases and color deficiency
- Animal and human trials are underway

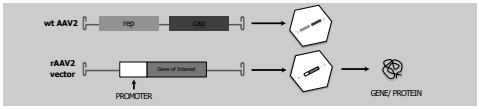


49

Adeno-Associated Virus (AAV) as a Gene Therapy Vector

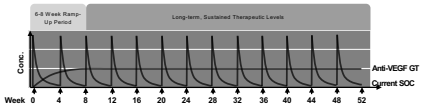
AAV

- Simple virus made safe for gene therapy
- Protein on outside, DNA on inside
- Non-pathogenic, non-replicating, non-integrating



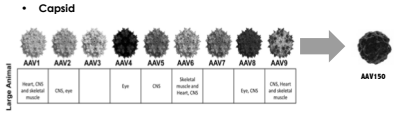
Sustained Delivery is "Holy Grail"

- Compliance with treatment is required for efficacy
- Gene therapy offers the potential for sustained and efficacious anti-VEGF protein delivery

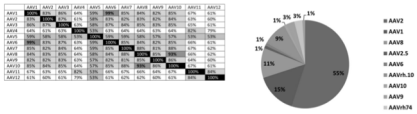


rAAV Capsid Variants

- Capsid

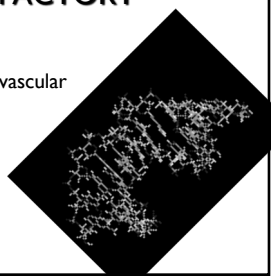


Summary of Trials to 2015



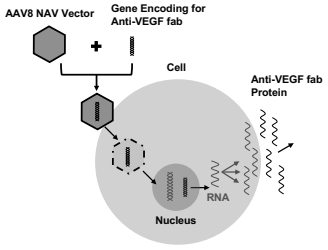
Gene Therapy
AMD ANTI-VEGF FACTORY

- RGX-314 Gene Therapy Subretinal Delivery for the Treatment of Neovascular AMD (nAMD)



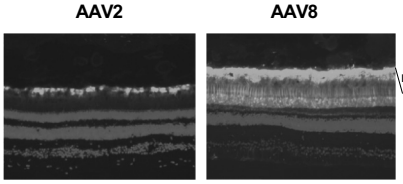
RGX-314 Optimized NAV Gene Therapy for Wet AMD

RGX-314 is Designed to Deliver a Gene Encoding for an Anti-VEGF fab Protein



53

RGX-314: Utilizing AAV8 for Higher Protein Expression in NHPs



More Efficient Gene Delivery to the RPE[1]

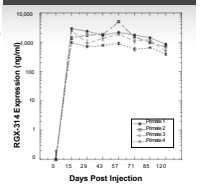
[1] Vandenberghe et al. 2011 *Science Translational Medicine*

54

RGX-314 Protein Expression in Aqueous Taps in NHPs

Sponsor	Vector	ROA	Transgene	Dose (GC/eye)	Max. Expression (ng/ml)[2]
REGENXBIO	AAV8	Subretinal	anti-VEGF fab	1.0e11	4,992
AVALANCHE	AAV2	Subretinal	sFlt	8.0e11	0.217
genzyme	AAV2	Intravitreal	sFLT01	2.4e10	528

RGX-314 Expression in Non-human Primates[1]



55

Sub-retinal vs Intravitreal for Gene Therapy

Sub-retinal:

- More invasive: surgical procedure
- Higher protein expression
- Immune privileged space- NAbs do not block transduction

56

RGX-314 Developing the Procedure and Training

Wet Lab and Training

57

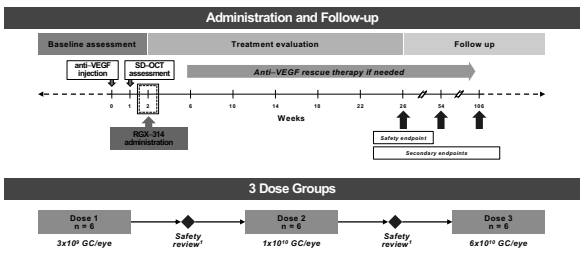
Standardized, controlled, automated sub-retinal injection

Fill Syringe

Sub-retinal Injection

RGX314 Subretinal delivery

RGX–314 Phase I Clinical Trial Design

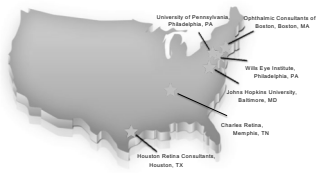


[1] Dose escalation safety review to occur four weeks after final subject in each cohort has been dosed

REGENXBIO

Acknowledgments

- Jeff Heier, MD (Boston, MA)
- Allen Ho, MD (Philadelphia, PA)
- Peter Campochiaro, MD (Baltimore, MD)
- Albert Maguire, MD (Philadelphia, PA)
- David Brown, MD (Houston, TX)
- Jorge Calzada, MD (Memphis. TN)
- Szilard Kiss, MD (New York, NY)
- Sherri Van Everen, PharmD (REGENXBIO)
- Stephen Yoo, MD (REGENXBIO)



REGENXBIO