Gene Therapy for Usher Syndrome (and other treatments)

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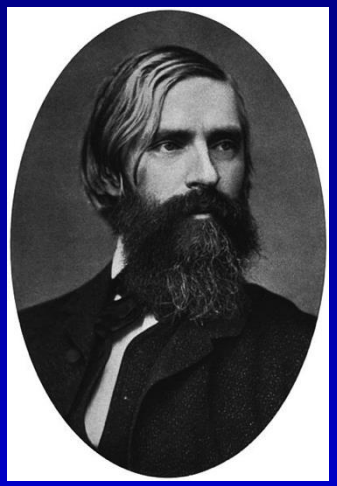
Usher Syndrome - History



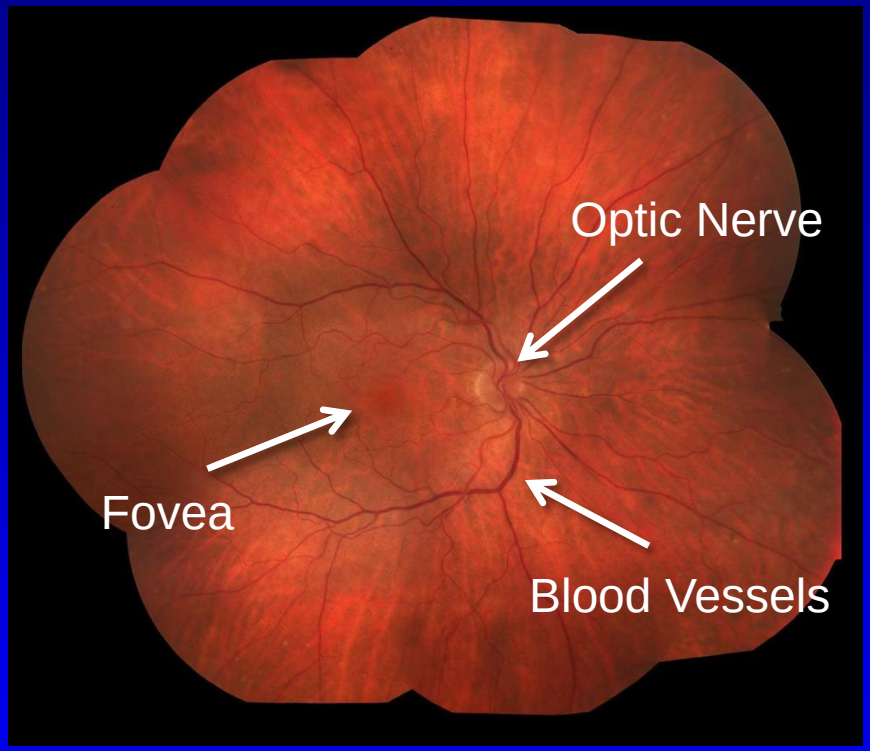
Albrecht von Gräfe

- First reported by Albrecht Von Graefe in 1858 where he described three brothers with deaf-blindness

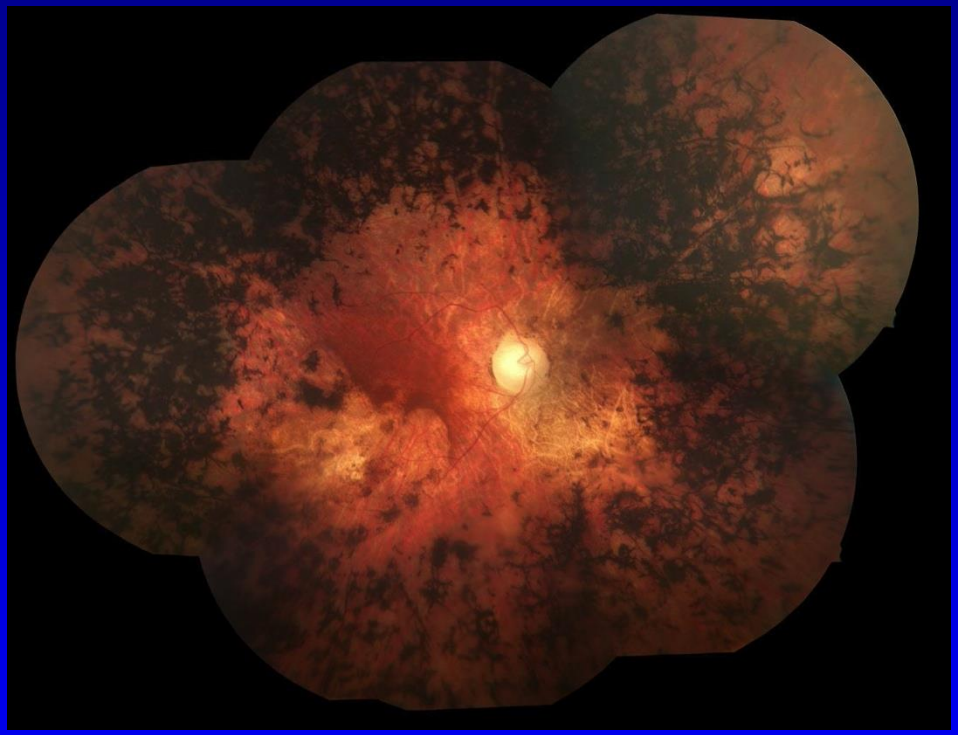
Charles Usher

- Scottish Ophthalmologist Charles Usher described 68 patients in 1912 with retinitis pigmentosa and deafness

Retinal Degeneration in Usher Syndrome



Normal Retina



Usher Syndrome

Anatomy of the Eye and Retina

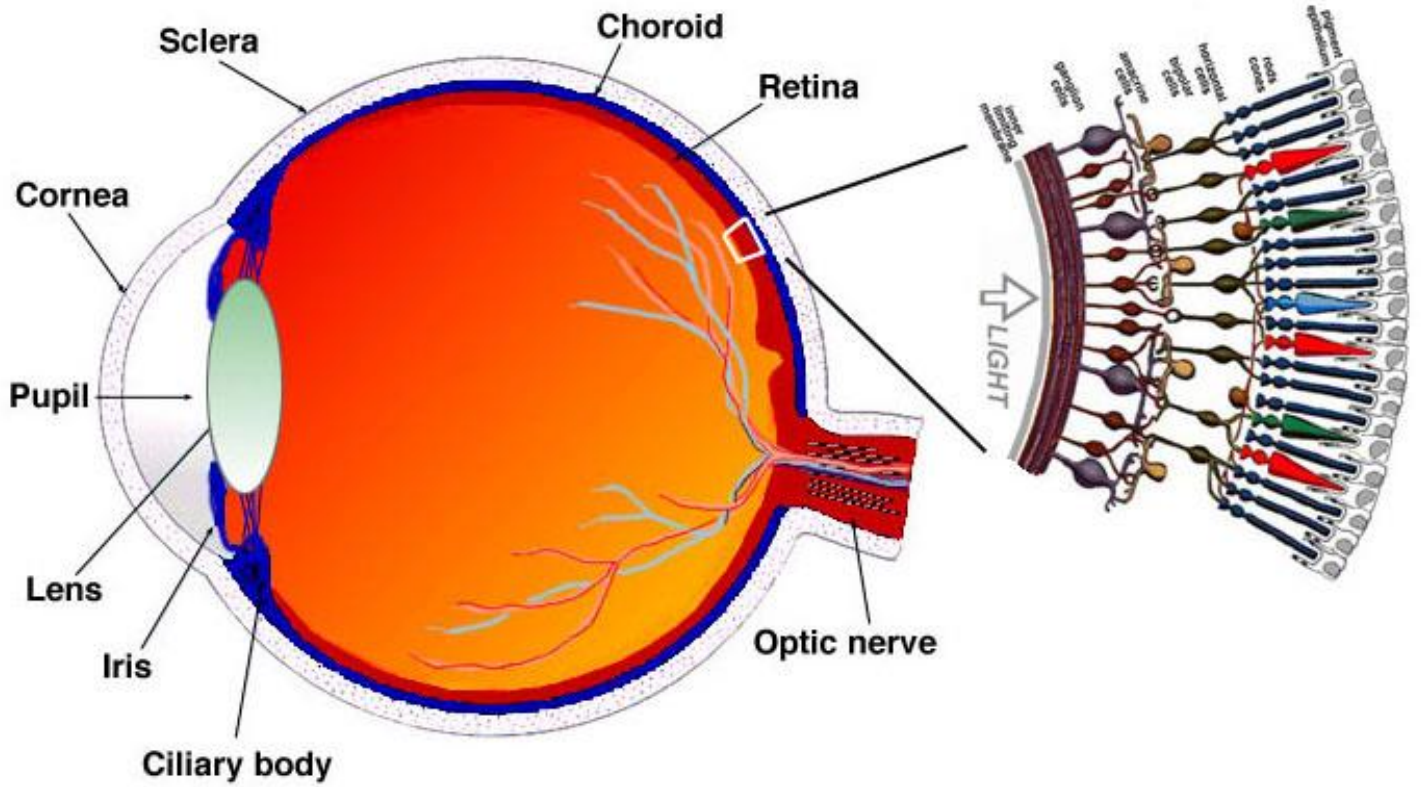
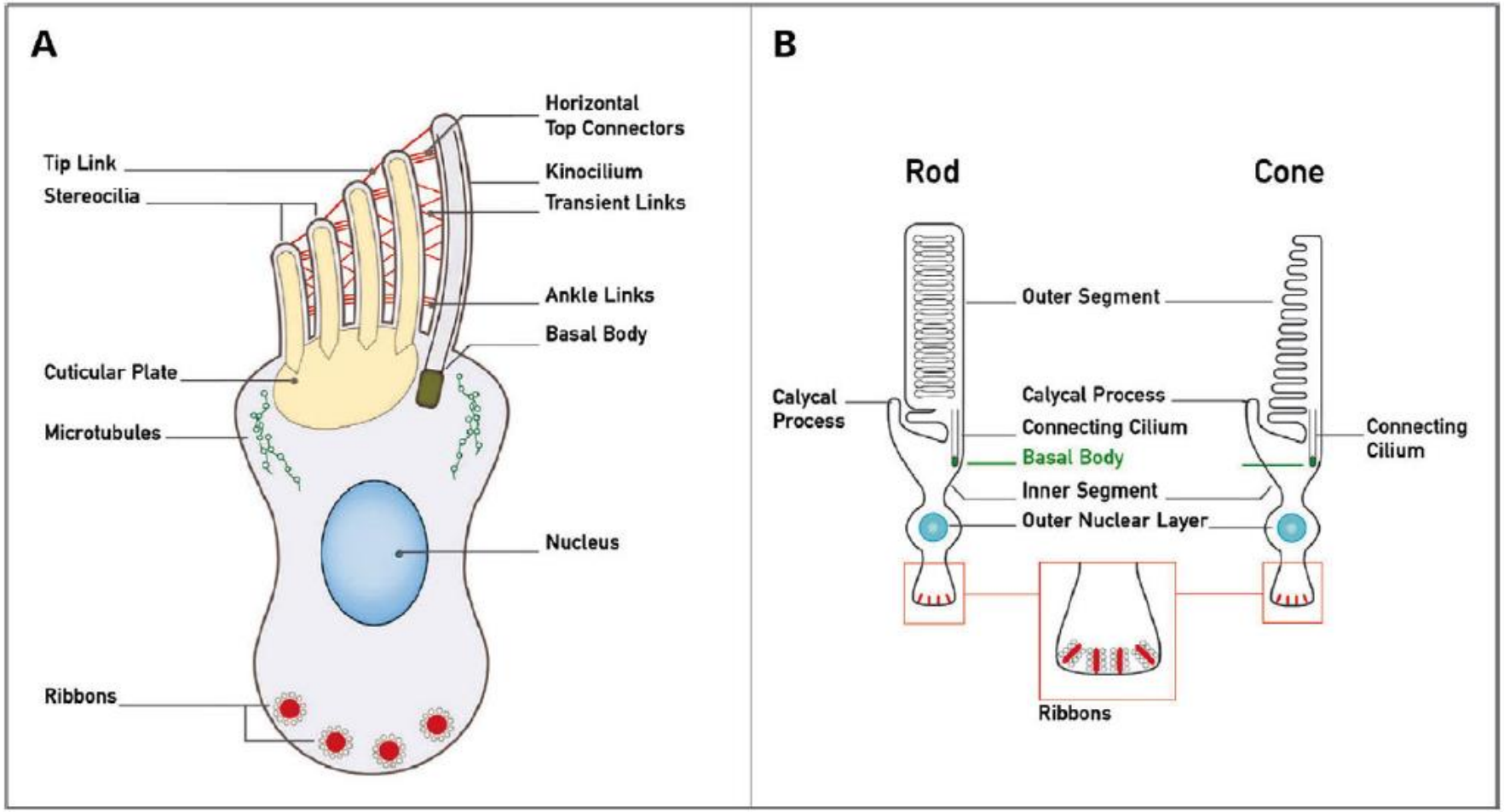


Fig. 1.1. A drawing of a section through the human eye with a schematic enlargement of the retina.

Images from Webvision

Rods and Cones die in inherited retinal degenerations

Hair cells and photoreceptors share specialized cilia



Usher Syndrome - Subtypes

Type I:

- Retinitis Pigmentosa – very severe
- Profound congenital deafness (cochlear implants)
- Vestibular dysfunction (balance problems)

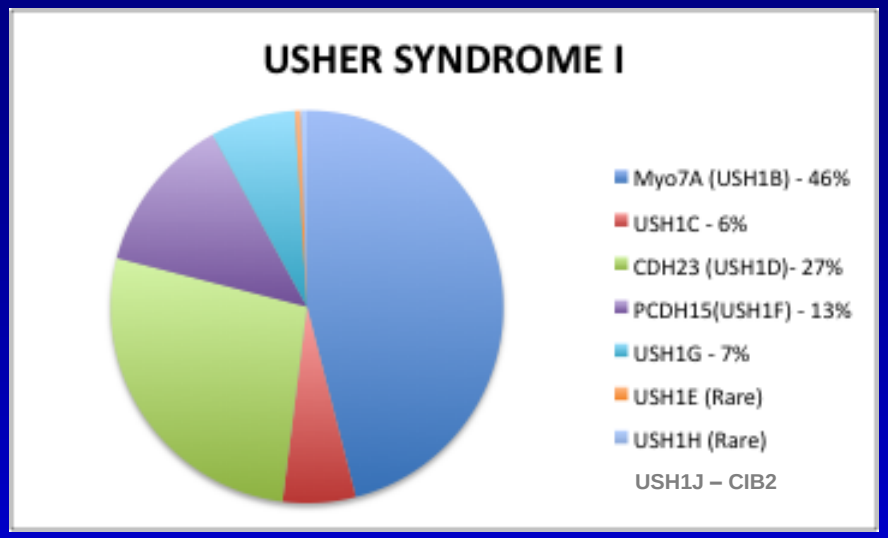
Type II:

- Retinitis Pigmentosa – moderate to severe
- Severe congenital deafness (hearing aids)

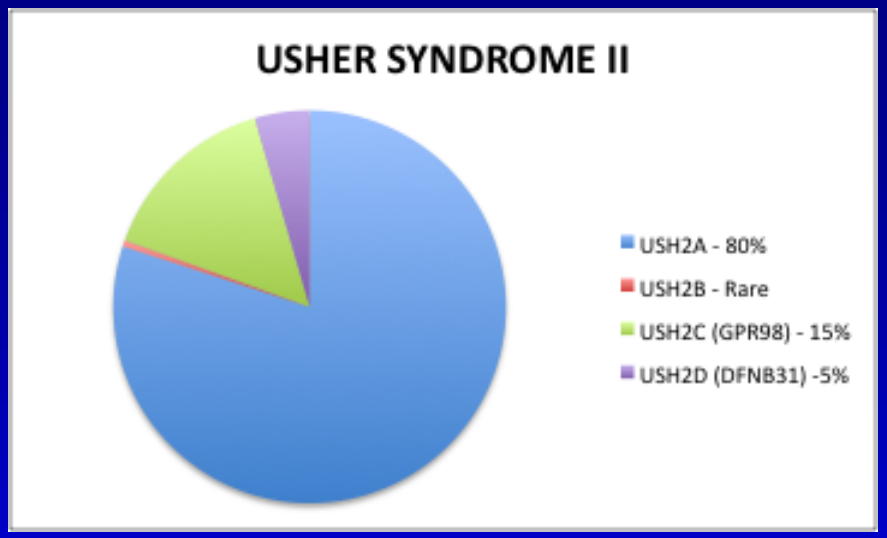
Type III:

- Retinitis Pigmentosa – moderate to severe
- Progressive deafness and vestibular dysfunction

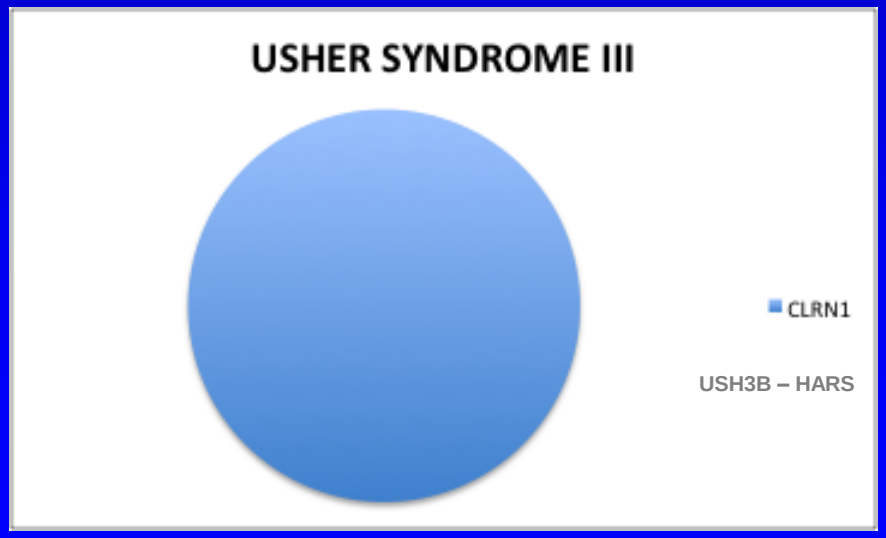
Genes Involved in Usher Syndrome



At least 8 genes

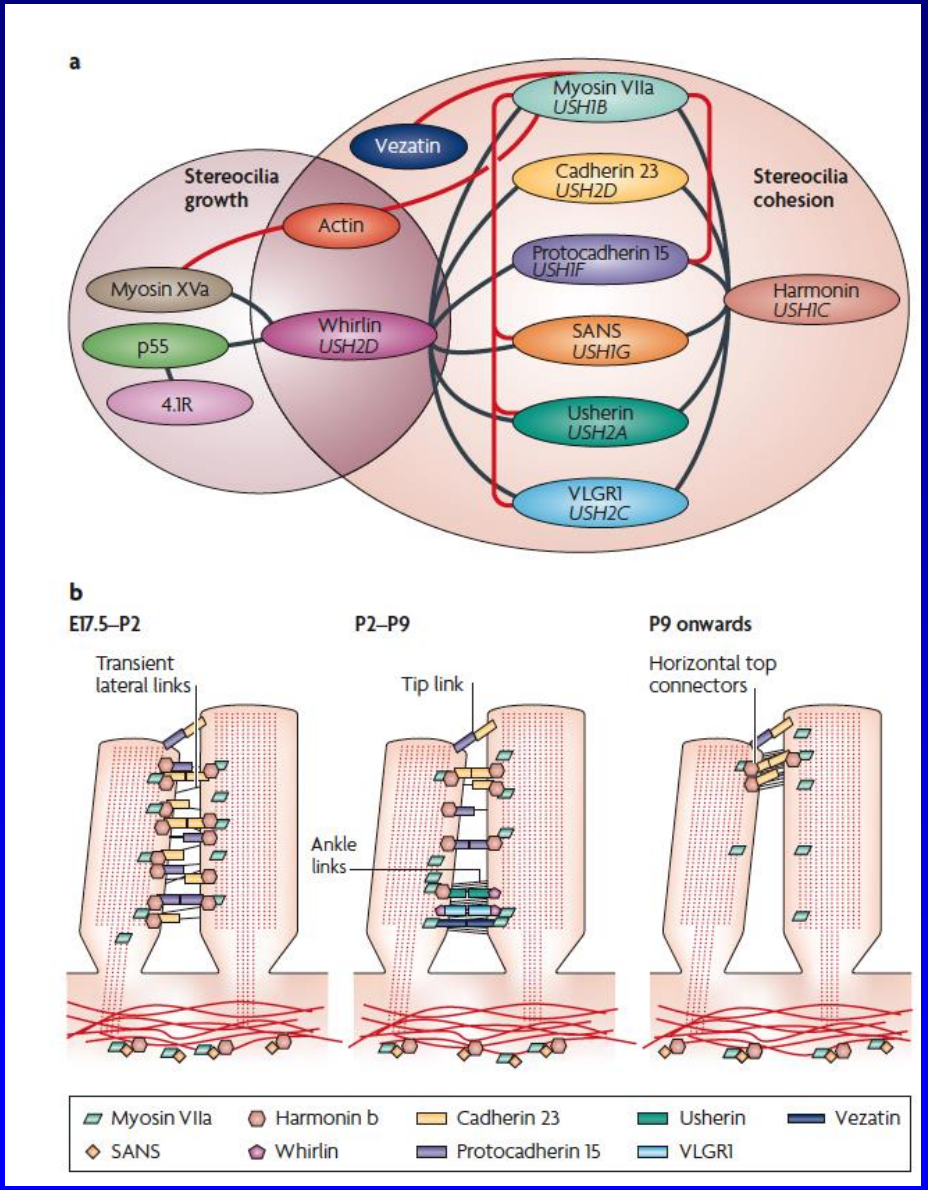


At least 3 genes



At least 2 genes

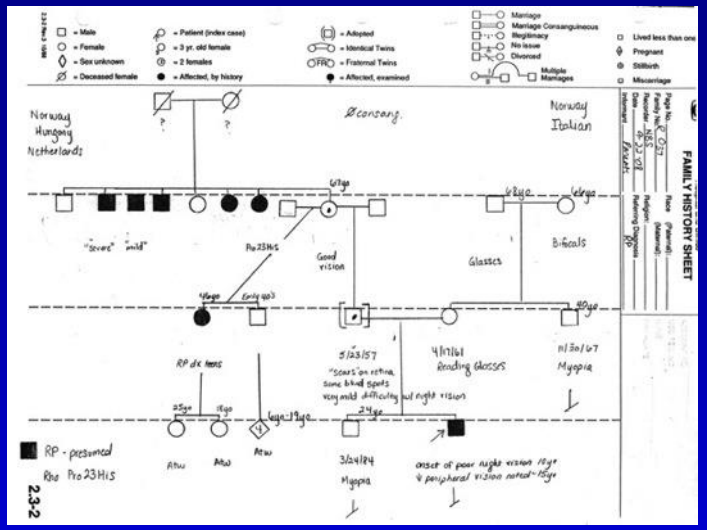
The Usher Interactome



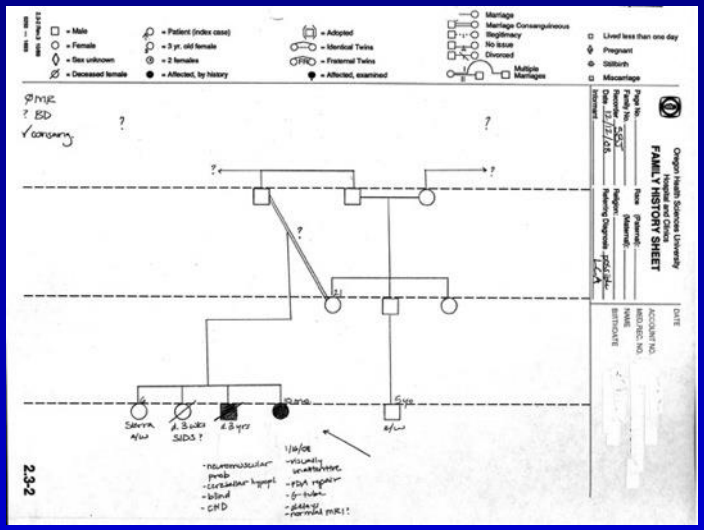
From Brown et al.

Tools to Study Usher Syndrome

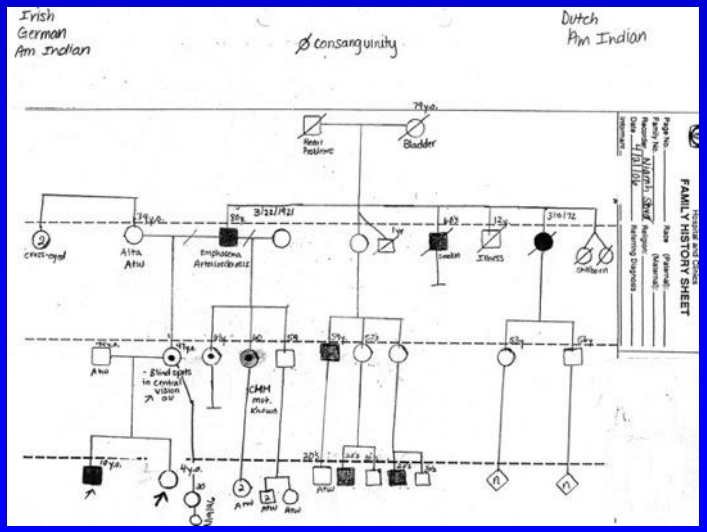
Family History



Autosomal Dominant



Autosomal Recessive

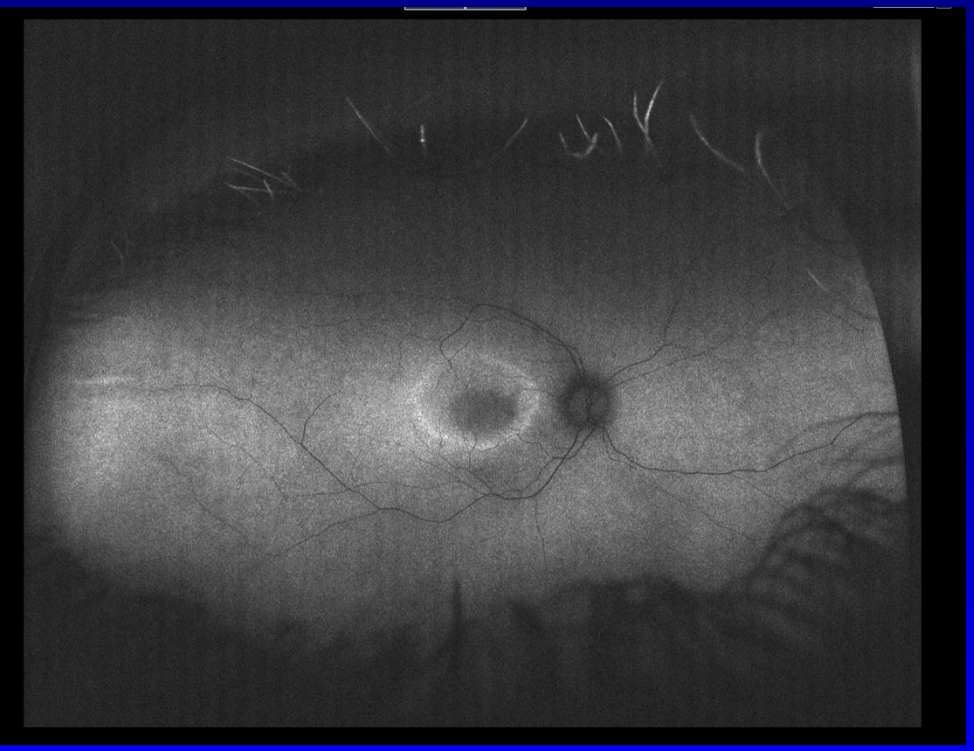


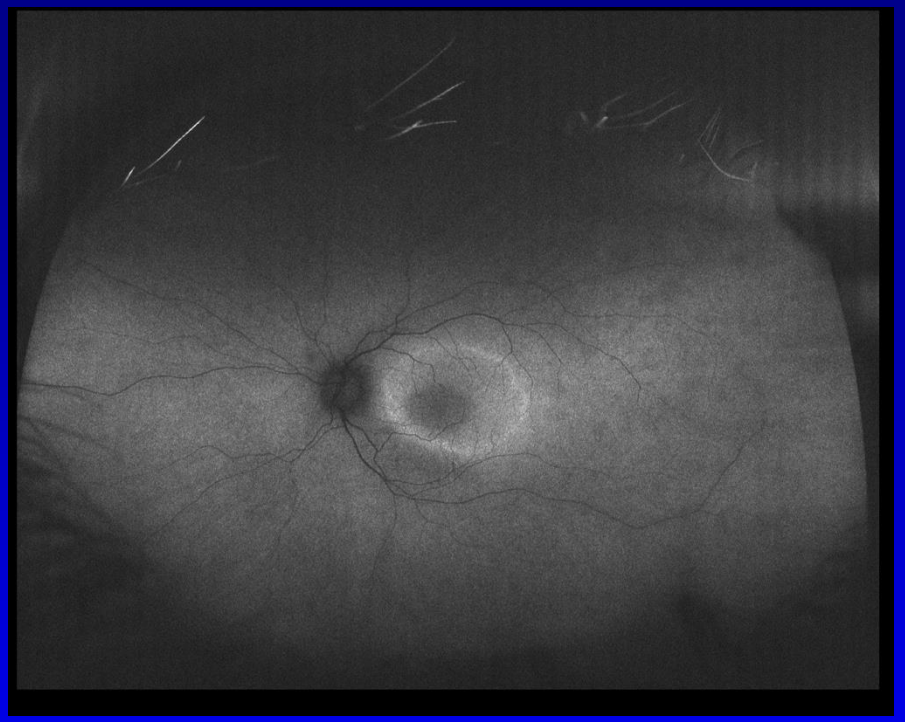
X-Linked Recessive

Fundus Photography

Patient with Type I Usher Syndrome

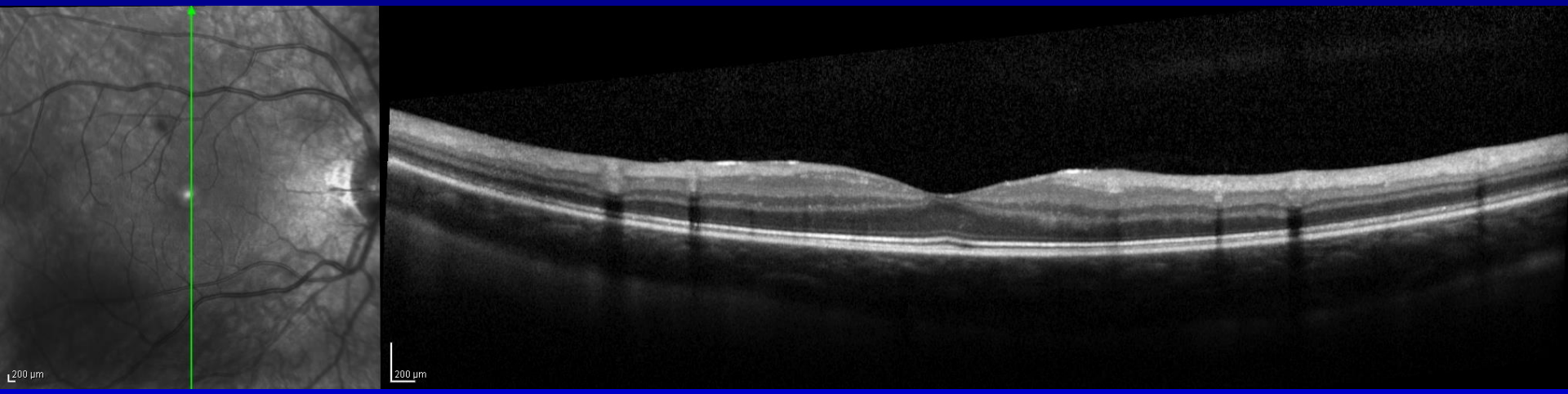
Short Wavelength Autofluorescence

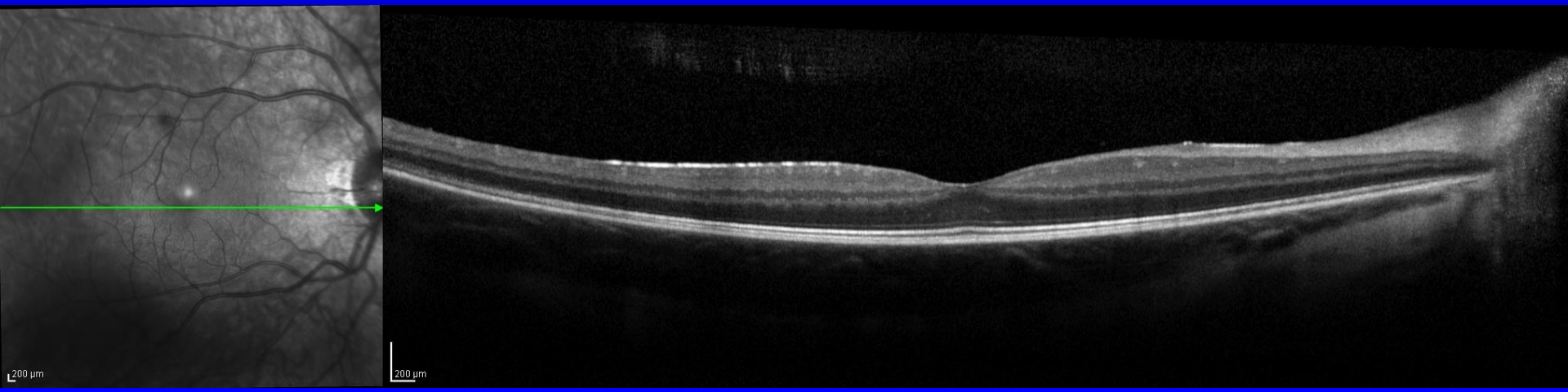




Patient with Type I Usher Syndrome

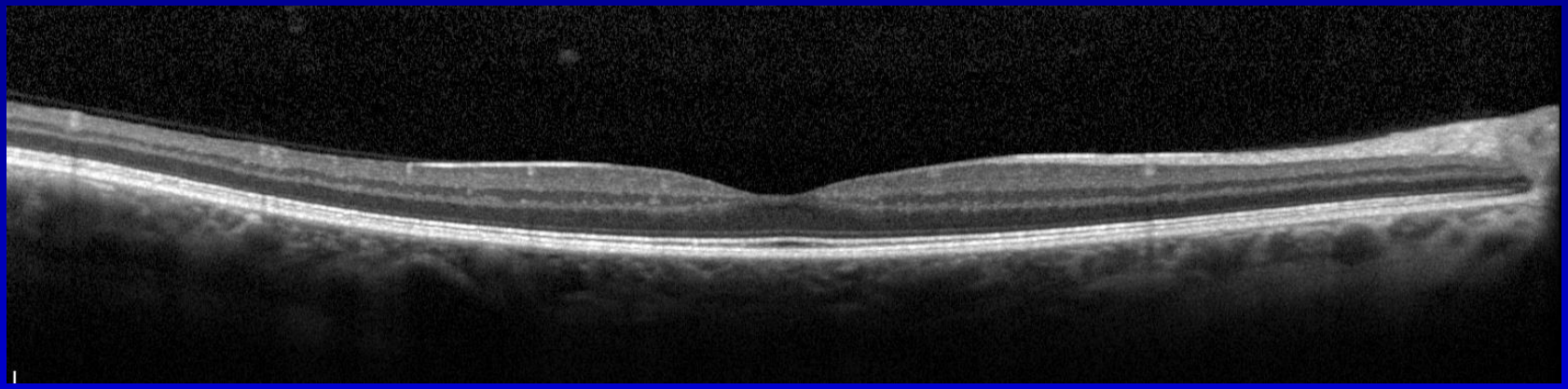
15 year old with USH1B from mutations of MYO7A



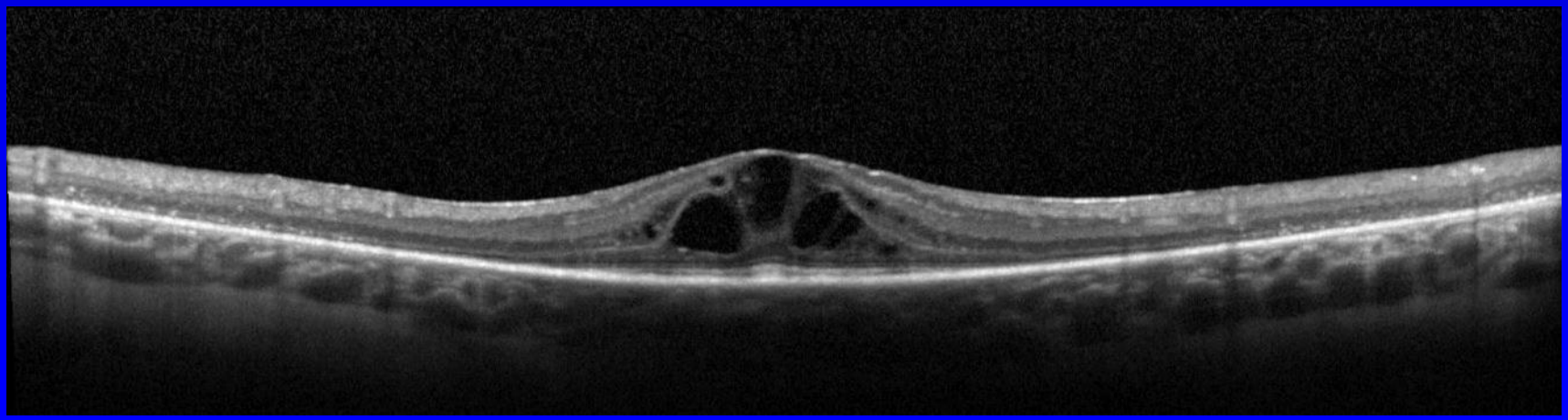


Importance of OCT for detecting Macular Edema

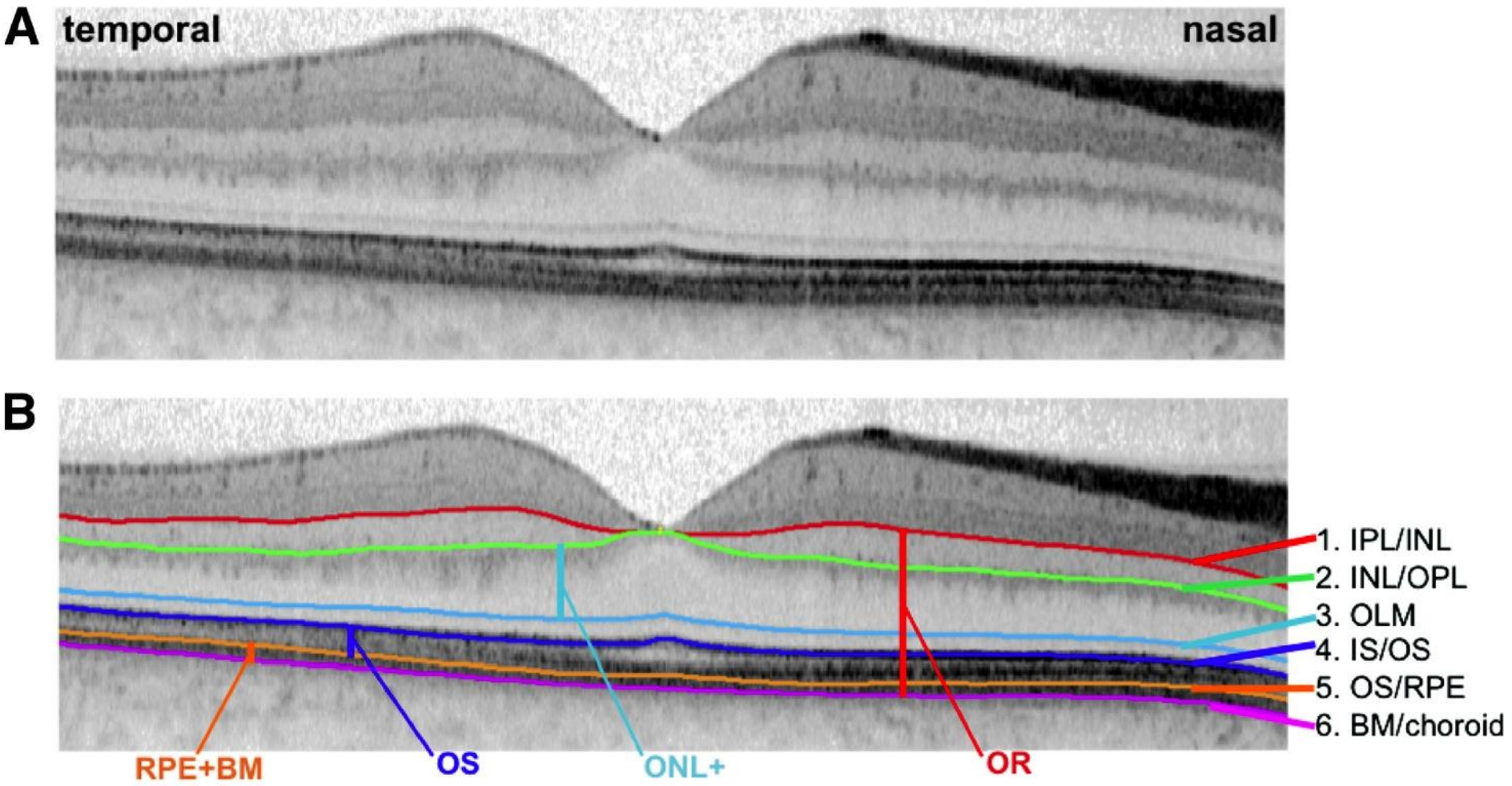
Healthy Eye



Cystoid Macular Edema in an Usher Patient

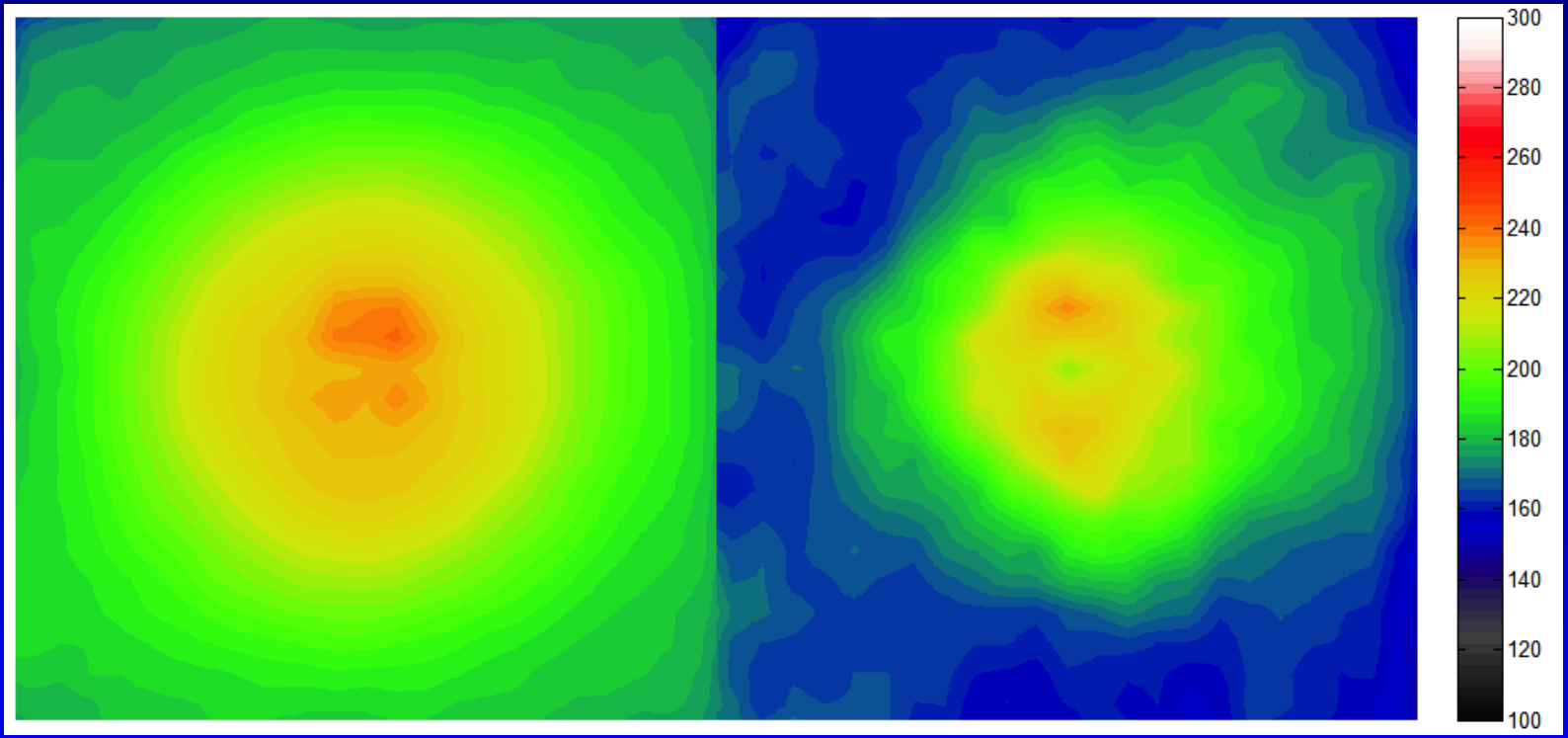


Segmentation of Retinal Layers



Lazow, MA, et al. Transition Zones between Healthy and Diseased Retina in Choroideremia (CHM), Stargardt Disease (STGD)Retinitis Pigmentosa (RP). Invest Ophthalmol Vis Sci 2011; 52: 9581

SD-OCT: Outer Retinal Thickness



Normal group AVG

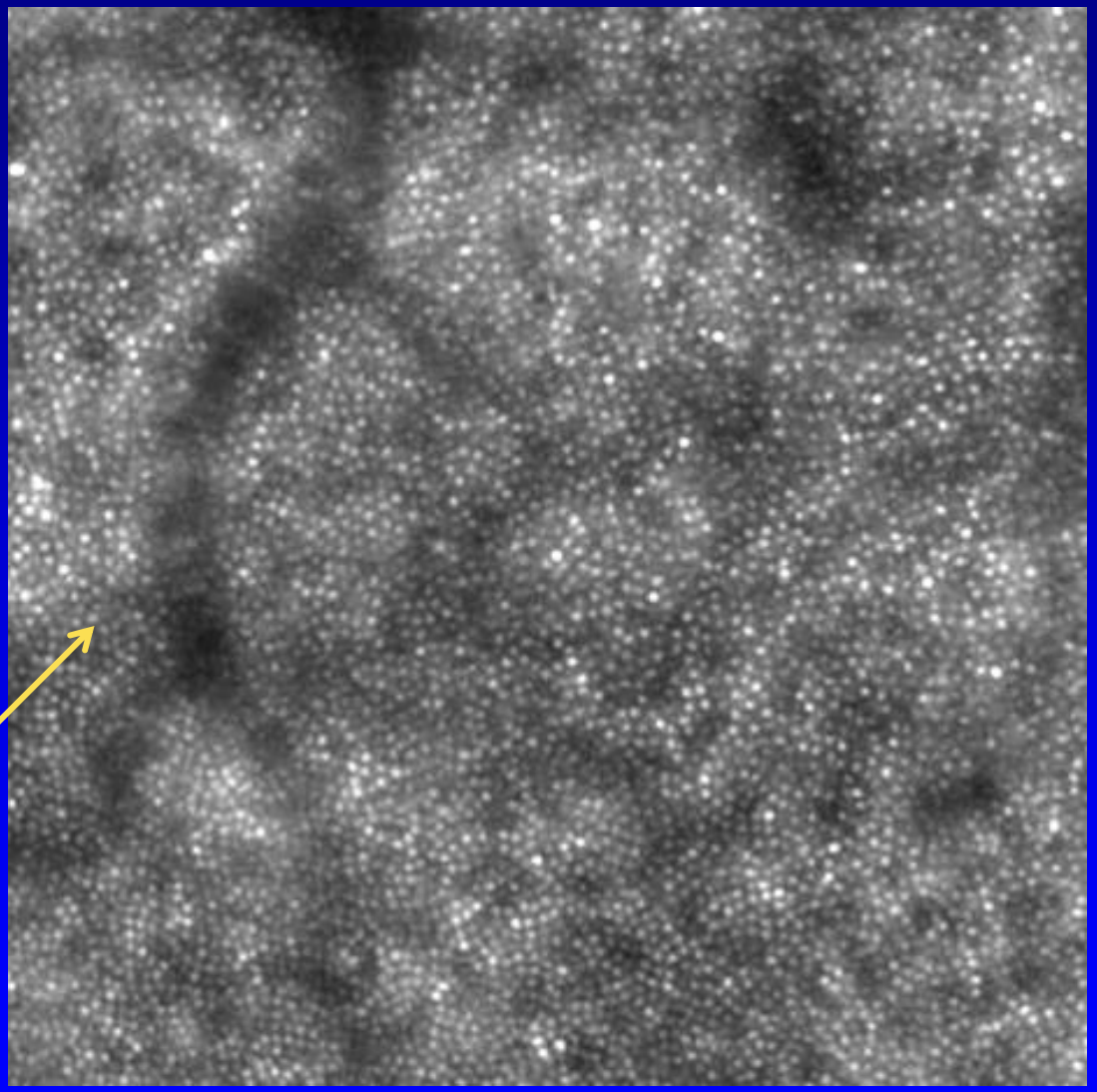
AMD group AVG

Overall average	Normal (28 Eyes)	AMD (12 Eyes)
Total retinal thickness(μm)	307.6±12.8	271.3±11.2
ORL thickness(μm)	194.5±6.6	175.1±10.1

Adaptive Optics

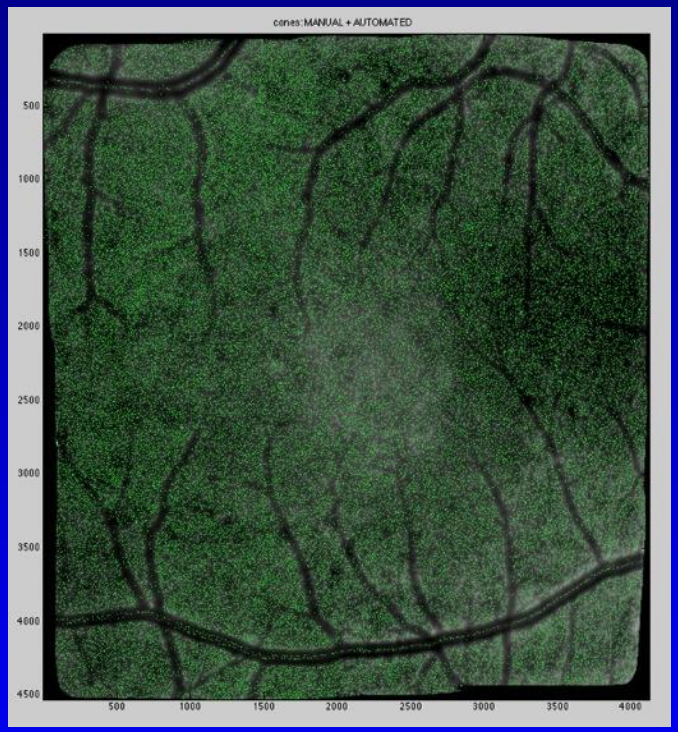
RTx1 from Imagine Eyes

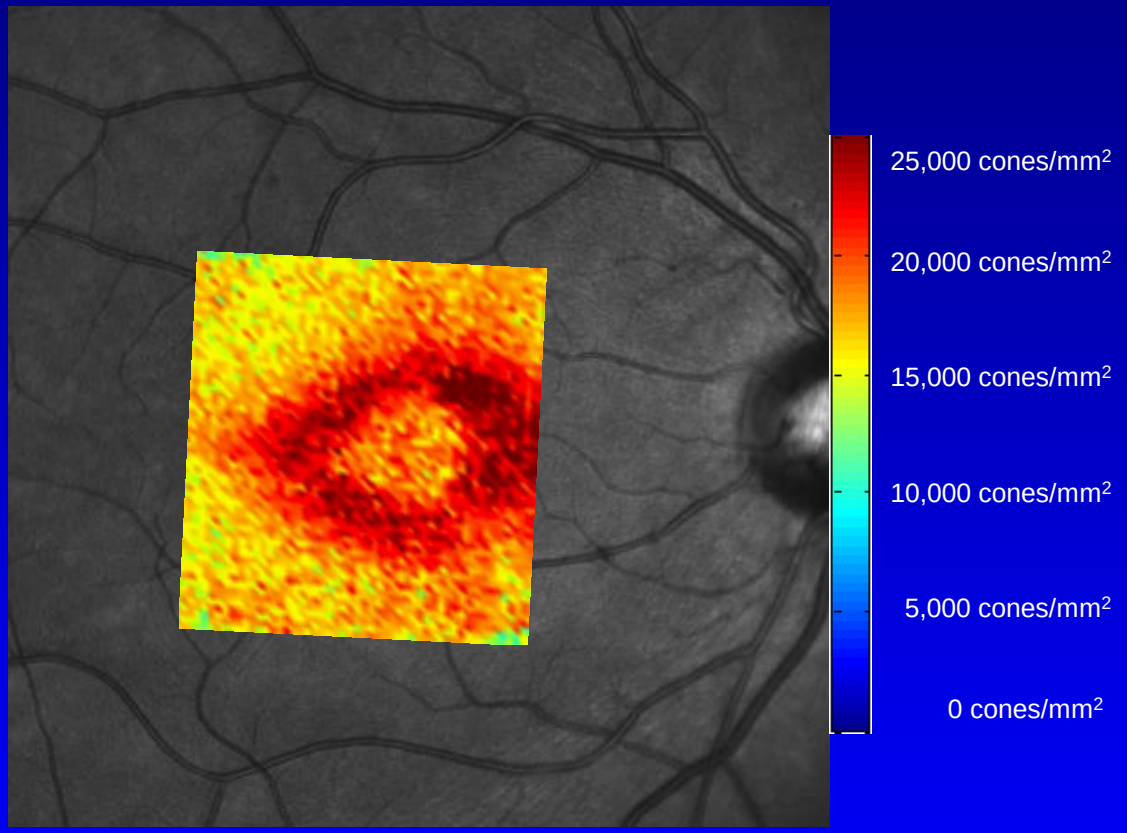
Individual Cone Photoreceptors



AO image demonstrating normal cone mosaic

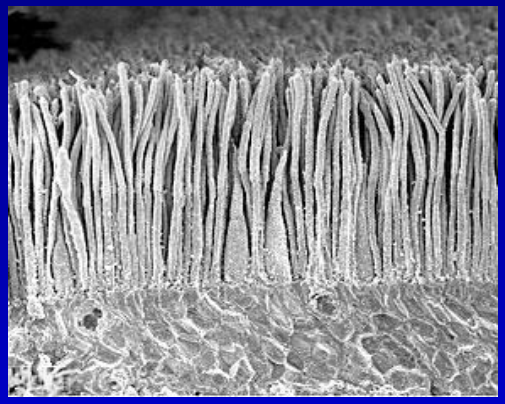
Adaptive Optics





Cone Density Map

Full Field Electroretinograms (ERG)



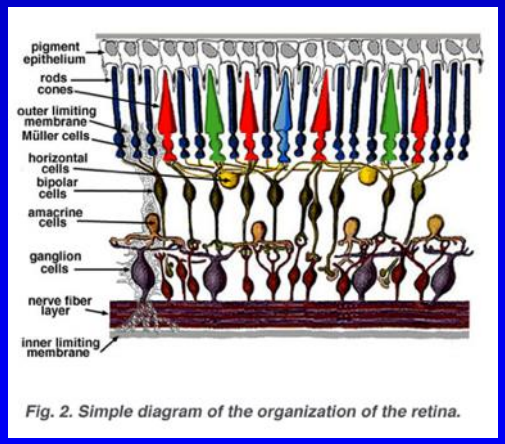
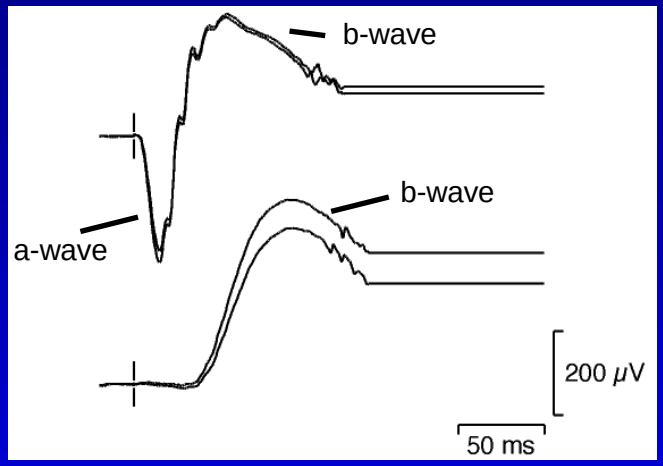
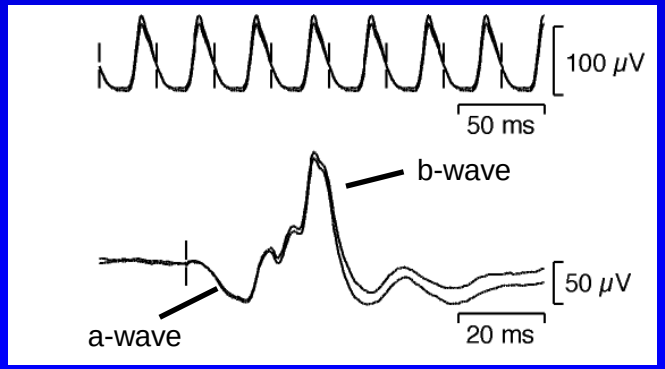


Fig. 2. Simple diagram of the organization of the retina.

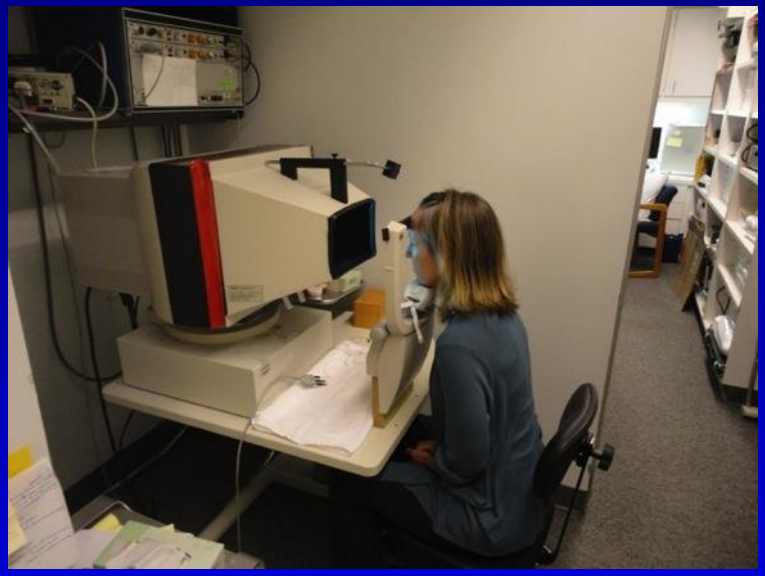
Rod-Driven ERG

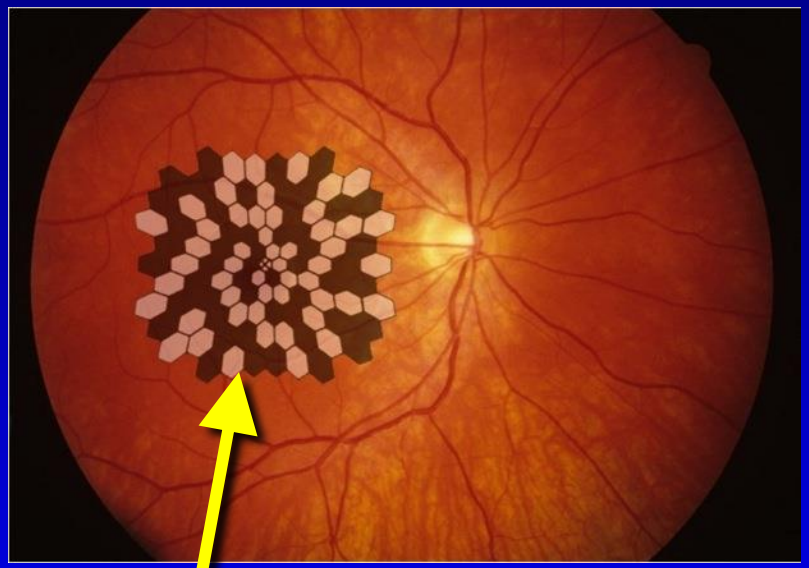


Cone-Driven ERG



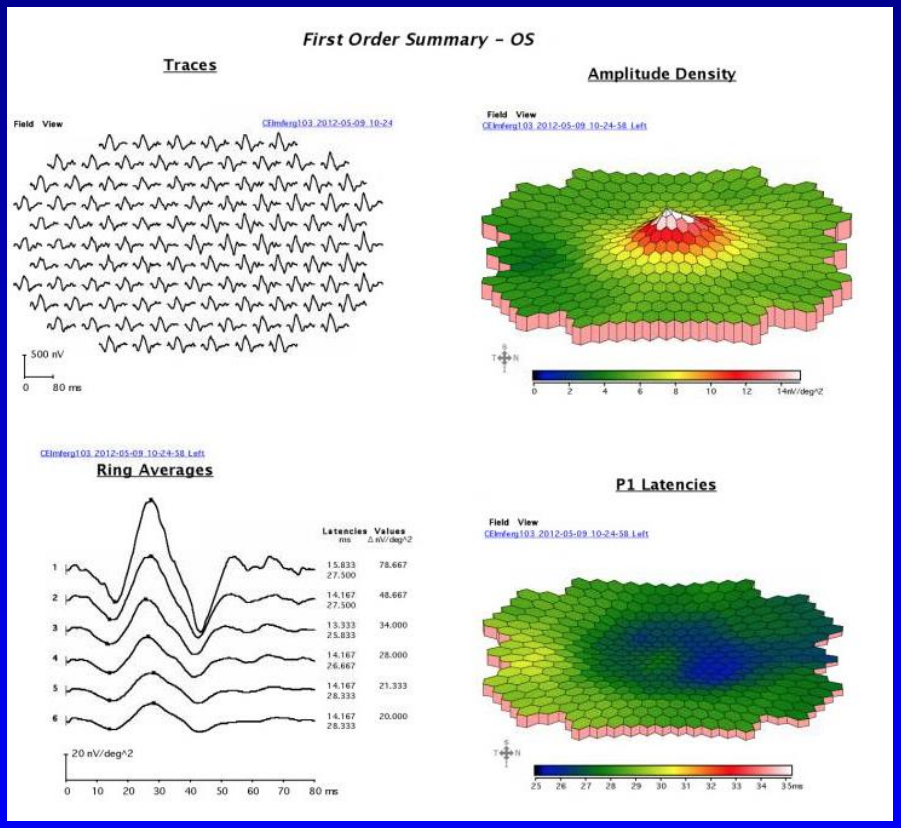
Multifocal Electroretinograms



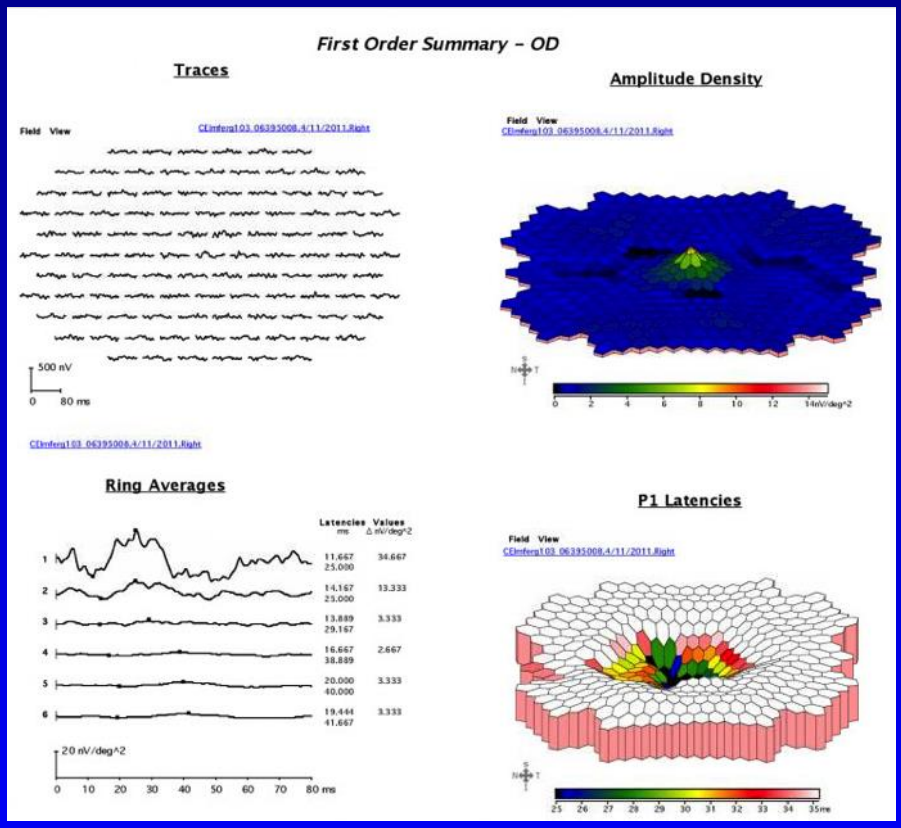


The mfERG only measures 24 deg!

Multifocal Electroretinograms



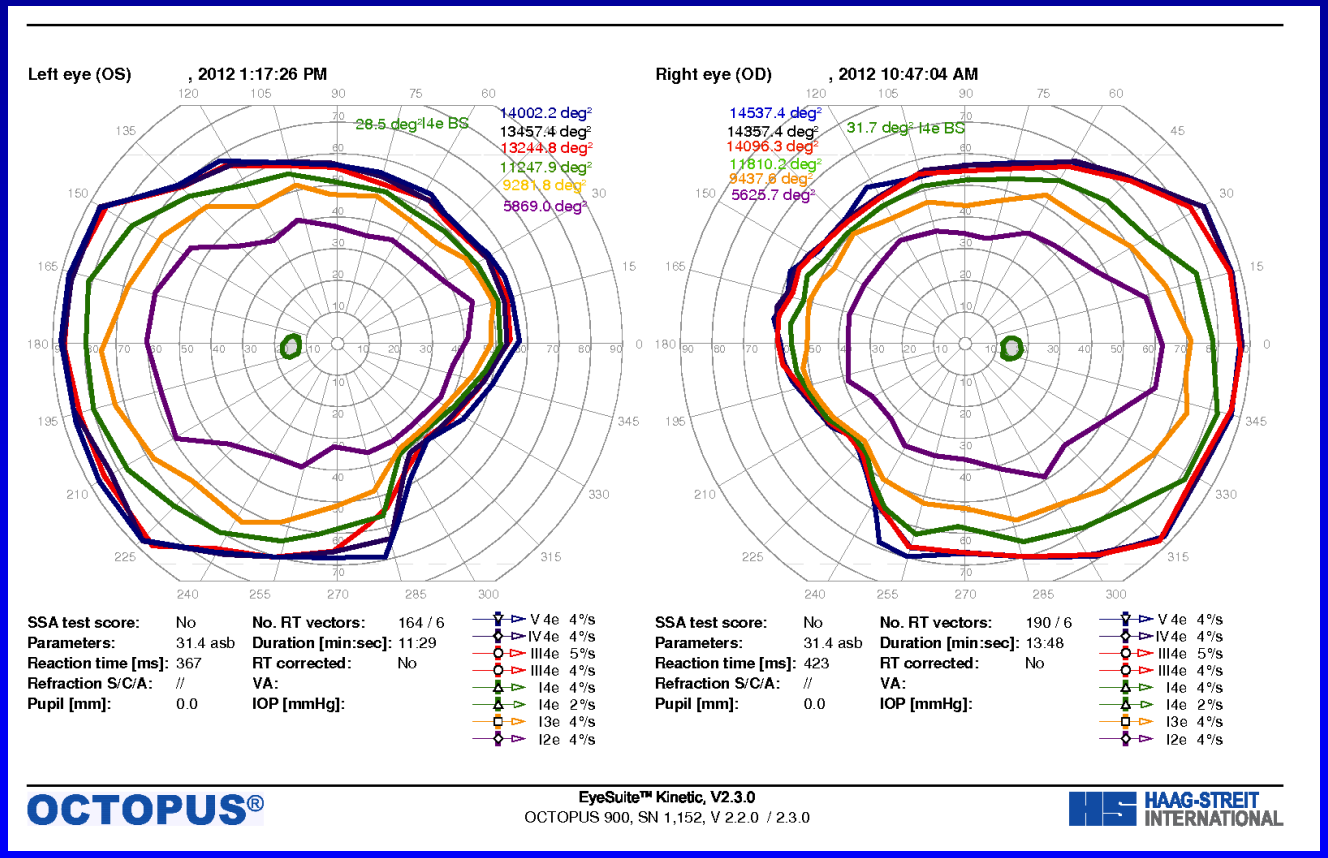
Healthy Eye



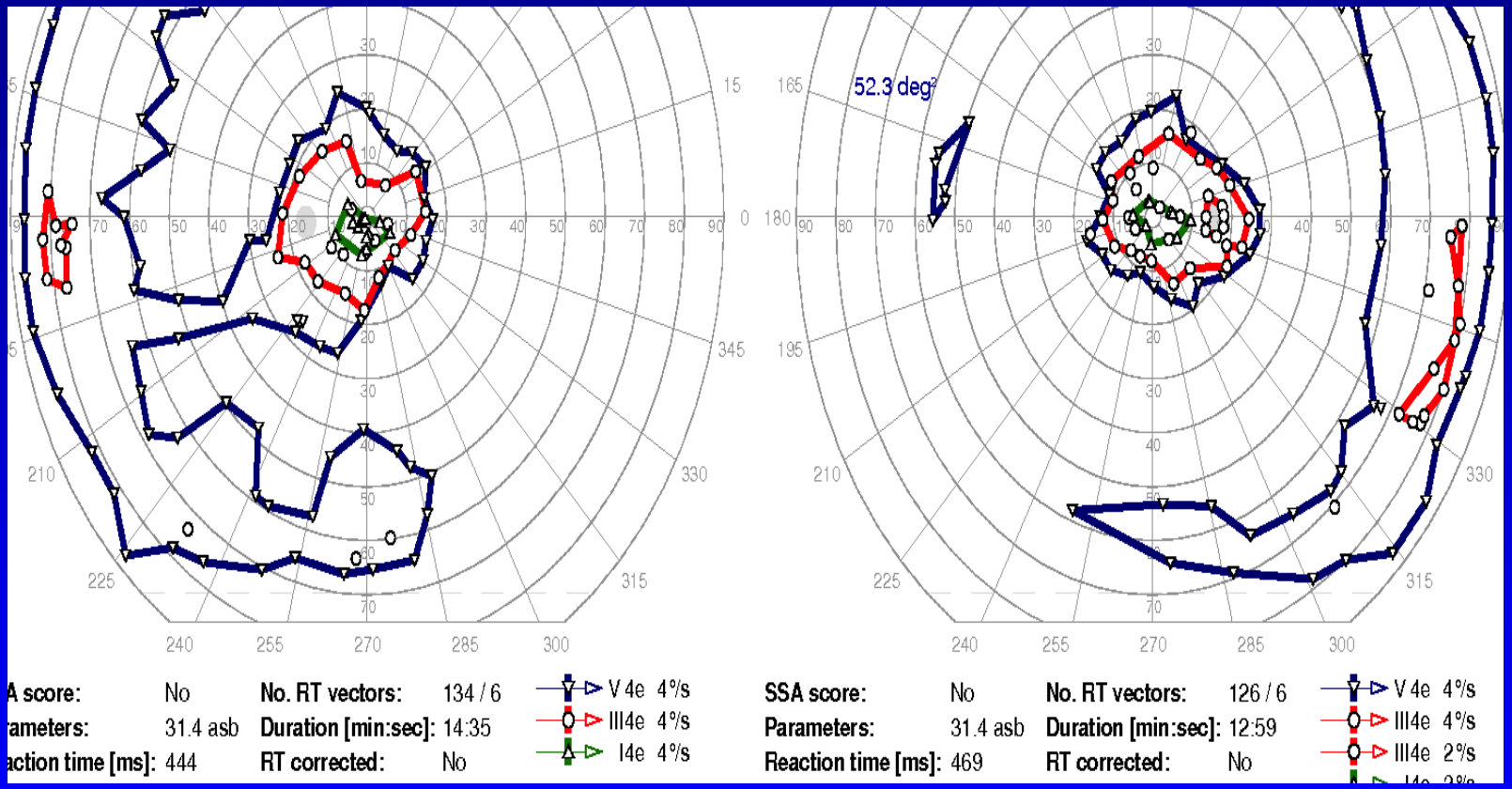
Usher Syndrome

Octopus Visual Fields

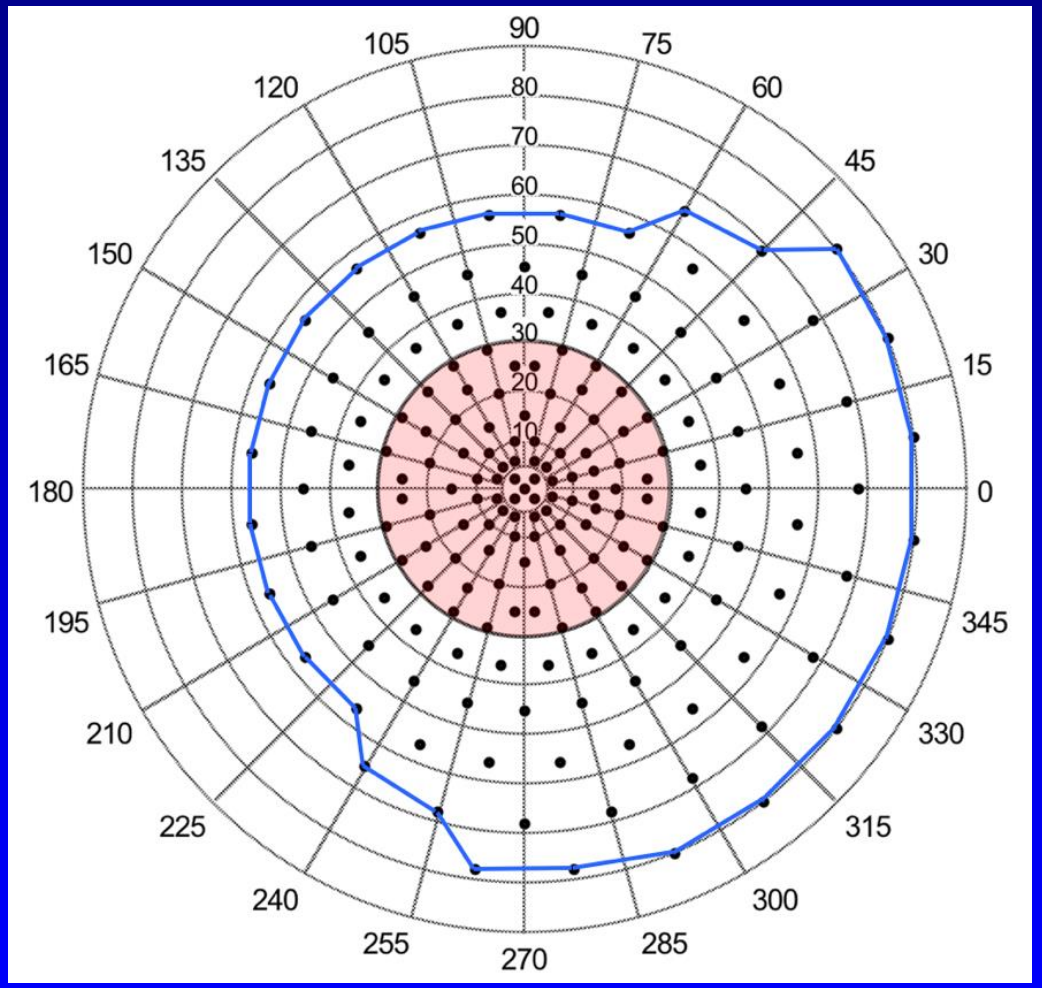
Kinetic Field



Kinetic Perimetry 24yr with USH1B



Faster GATE Algorithm allows 184 points to be sampled and cover entire field



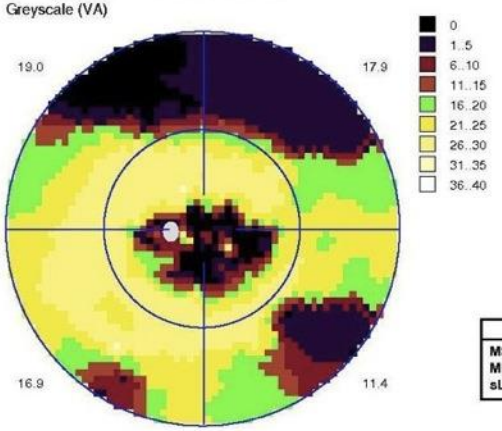
187 pt Grid

Octopus Visual Fields

Abnormal Static Field

ID

STGD-RP 184n GATE V
Left eye (OS) / / /2012 / 08:43:06
Greyscale (VA)

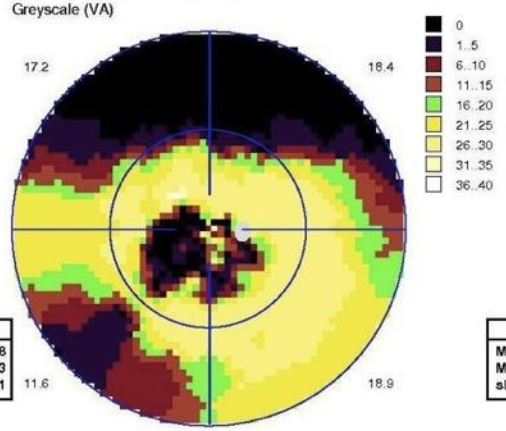


90°	
MS [dB]:	15.8
MD [< 2.0 dB]:	14.3
sLV [< 2.5 dB]:	13.1

Programs: CT Standard White/White / GATE
Parameters: 31.4 / 4000 asb V 200 ms
Catch trials: 1/33 (3%) +, 9/33 (27%) -
Refraction S/C/A: //
Pupil [mm]:
Questions / repetitions: 662 / 1
Duration: 25:15
RF: 15.1
VA:
IOP [mmHg]:

Comment:
Classification:

STGD-RP 184n GATE V
Right eye (OD) / / /2012 / 07:58:54
Greyscale (VA)



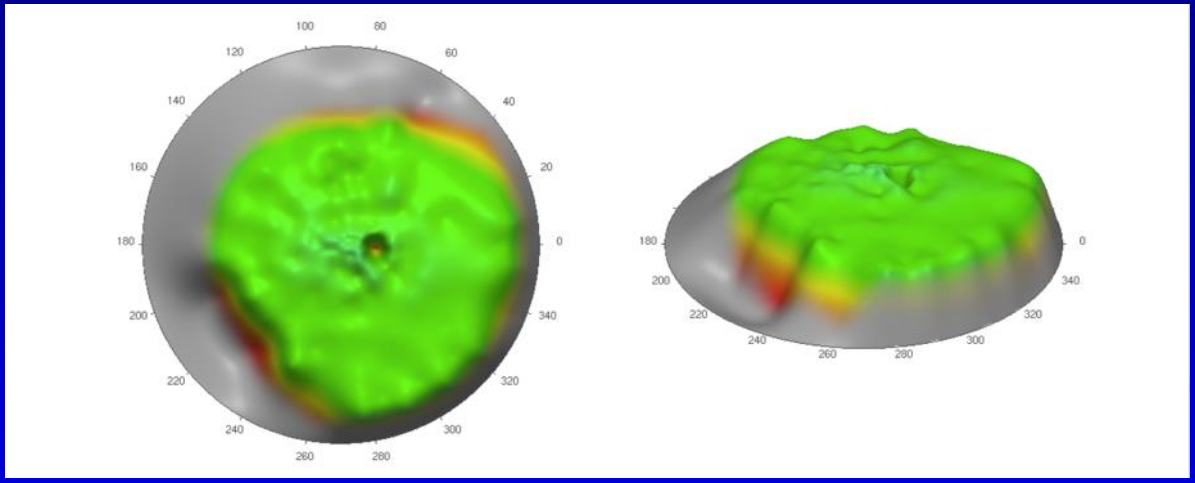
90°	
MS [dB]:	16.5
MD [< 2.0 dB]:	13.6
sLV [< 2.5 dB]:	12.7

Programs: CT Standard White/White / GATE
Parameters: 31.4 / 4000 asb V 200 ms
Catch trials: 3/32 (9%) +, 4/33 (12%) -
Refraction S/C/A: //
Pupil [mm]:
Questions / repetitions: 645 / 1
Duration: 24:13
RF: 10.7
VA:
IOP [mmHg]:

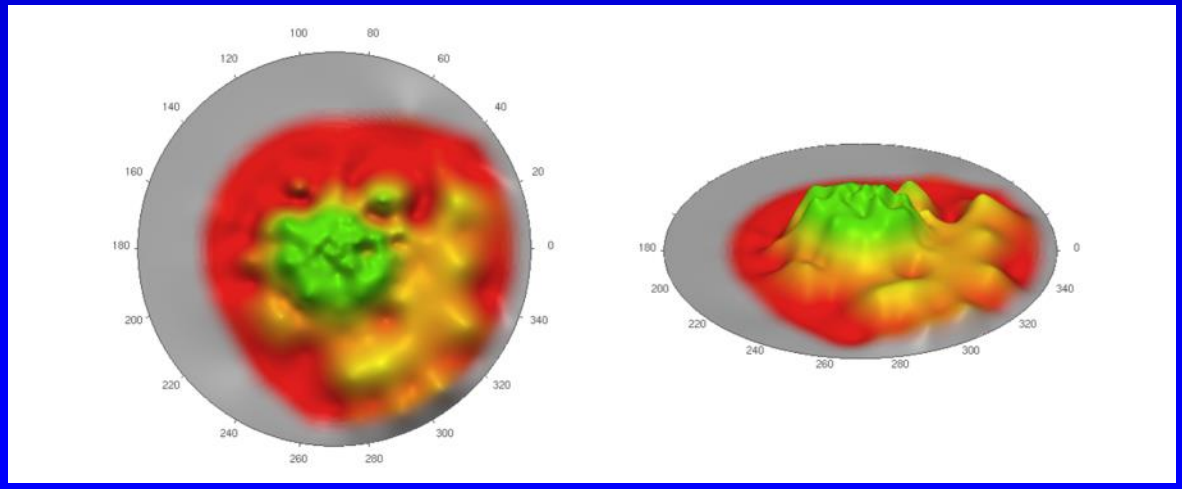
Comment:
Classification:

OCTOPUS®
EyeSuite™ Static perimetry, V2.3.0
OCTOPUS 900, SN 1152, V 2.2.0 / 2.3.0
HAAG-STREIT INTERNATIONAL

Visual Field Modeling in Usher Syndrome



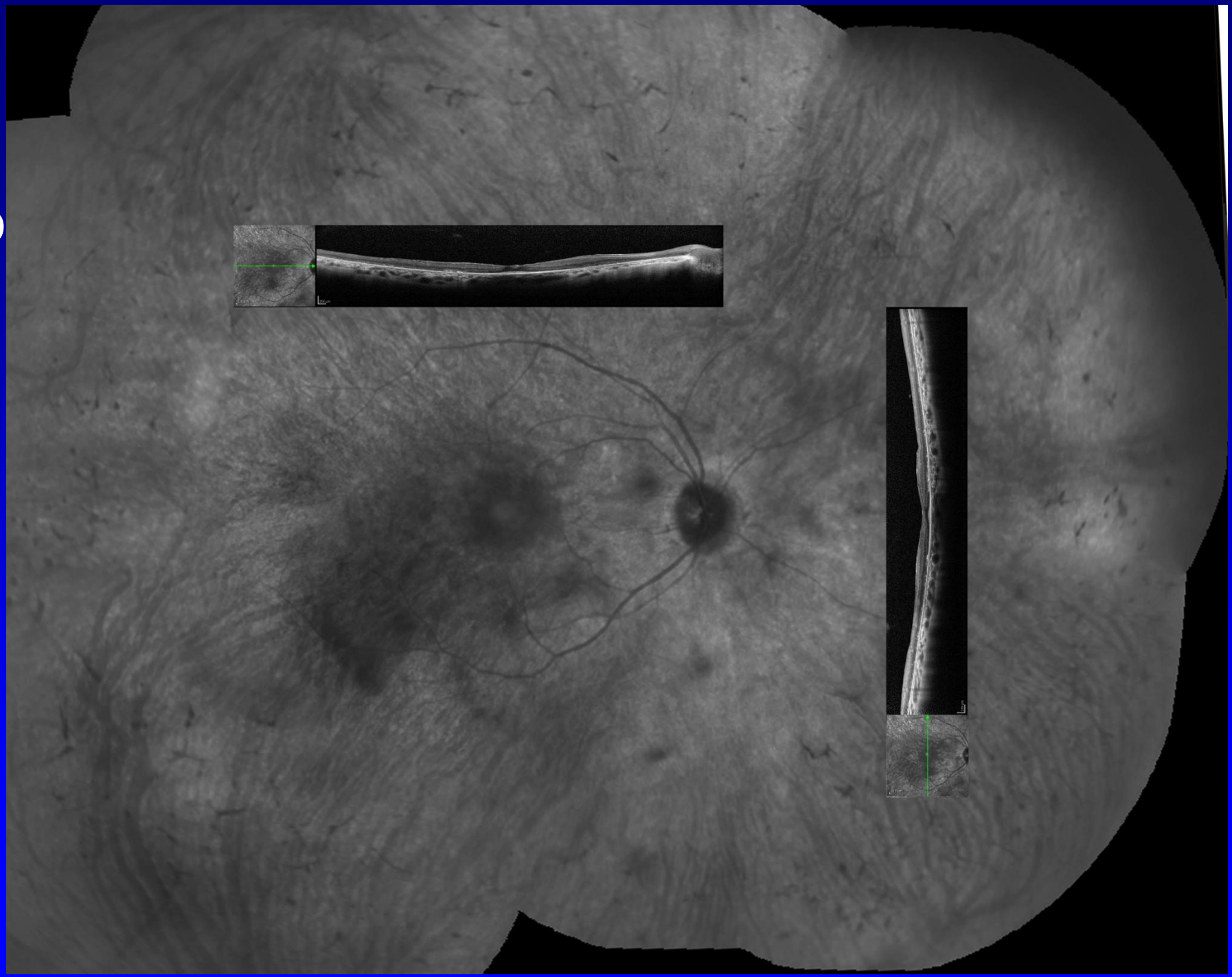
Normal

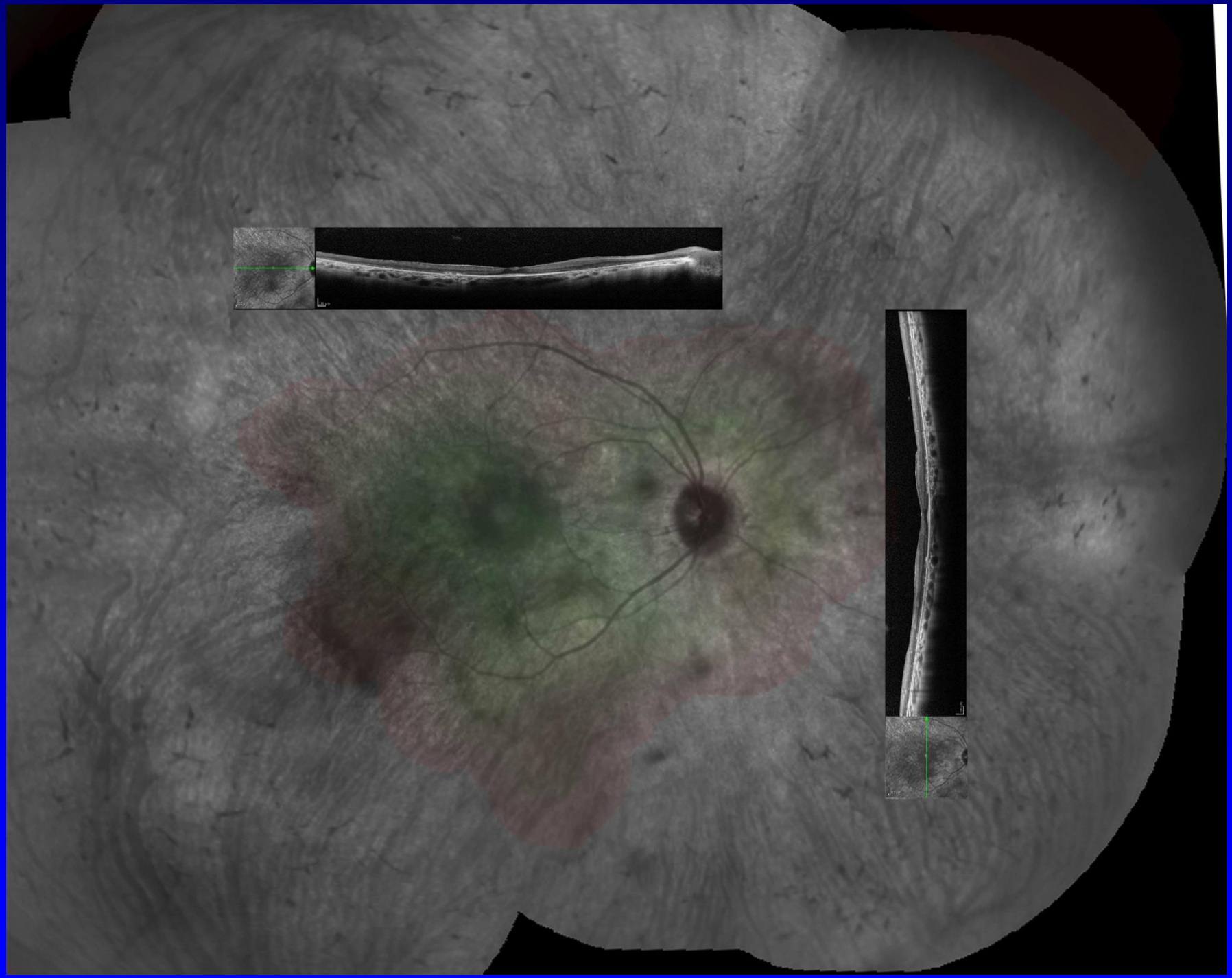


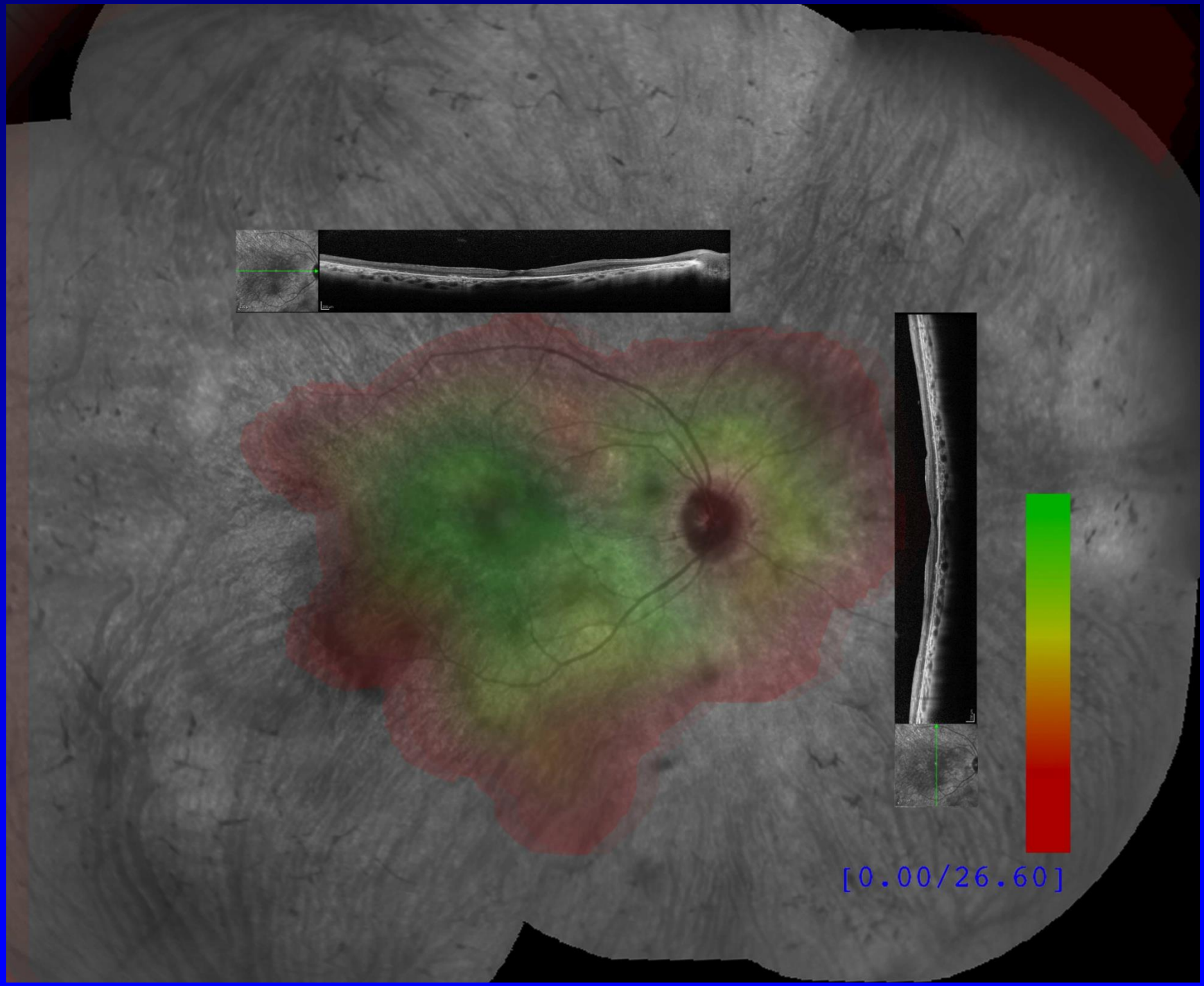
Usher Syndrome

Combining Structural and Functional Information

OD



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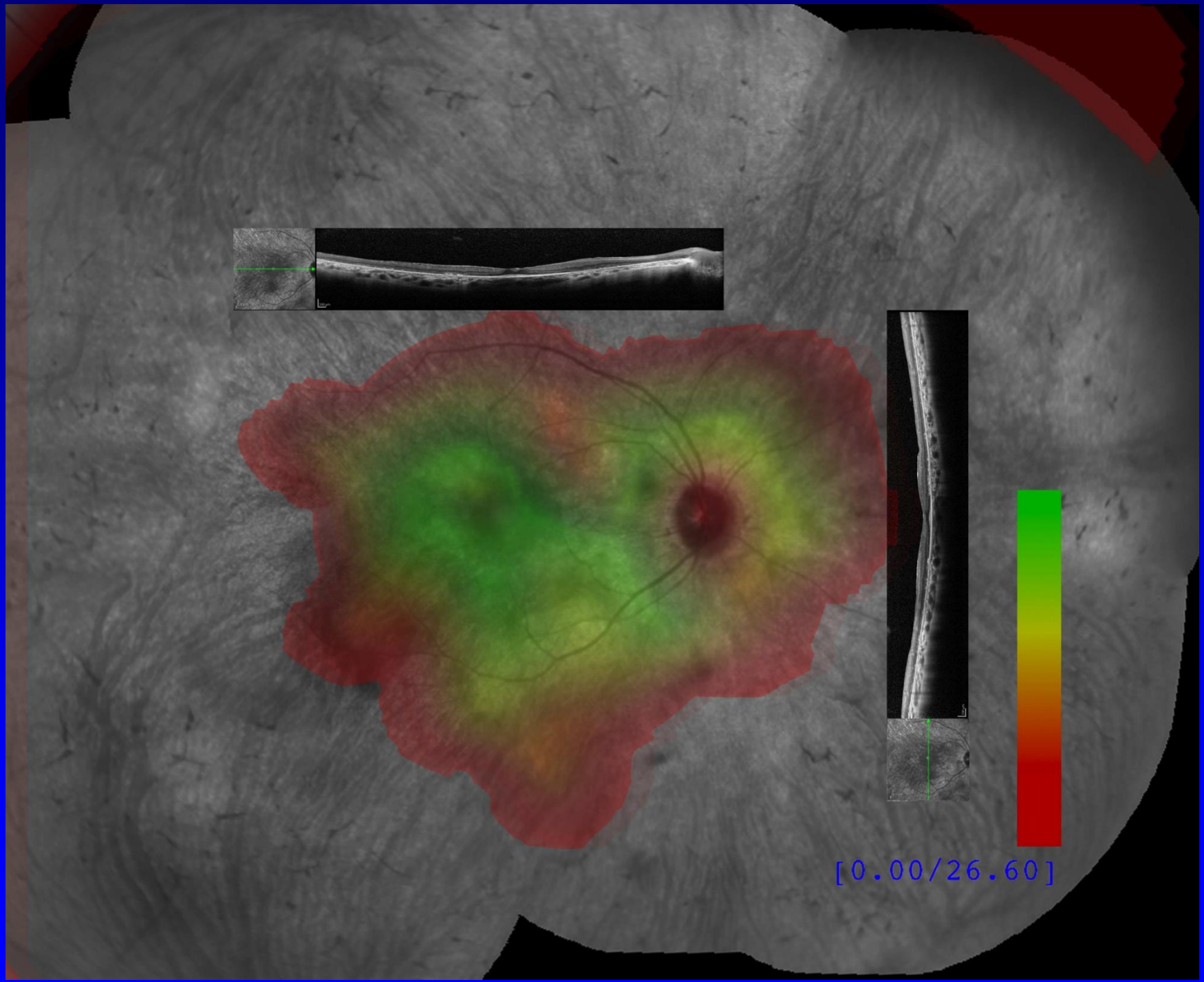
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Treatments for Retinal Degenerations

- Low Vision Aids
- Micronutrients
- Neuroprotection/Small Molecules
- Transcript Editing (Antisense Oligonucleotides)
- Cell Based Therapy (Stem Cells)
- Optogenetics
- Artificial Retina
- Gene Therapy

Low Vision Aids

Proper glasses prescription

Night vision goggles

Broad-beam Flashlight

Magnifiers and minifiers

Tablet Devices

Micronutrients and Vitamins

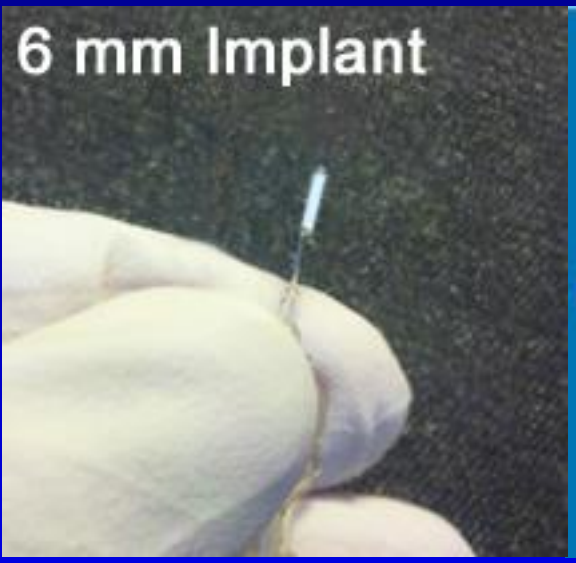
- High dose Vitamin A

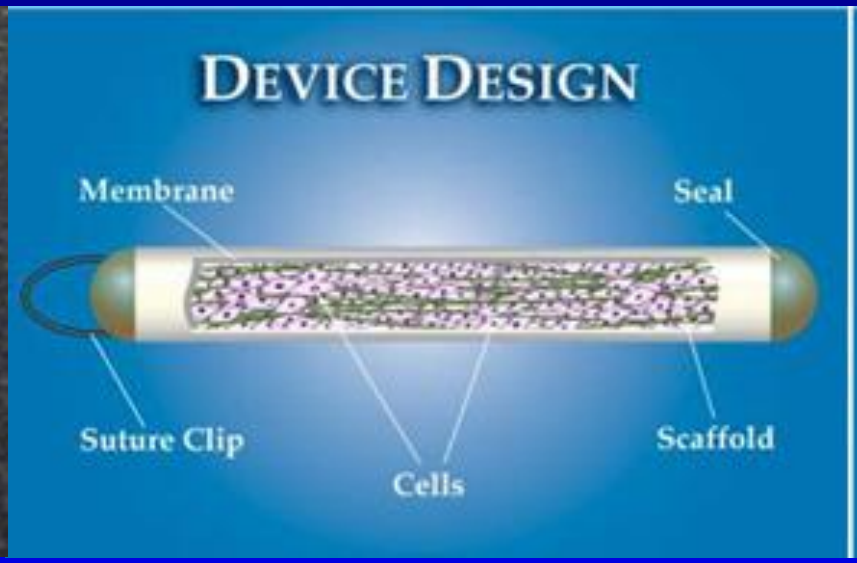
  Precautions for taking high doses of vitamin A
  - Liver Toxicity
  - Osteoporosis
  - Pregnancy
  - contraindicating in some mutations (such as those seen in Stargardt Disease)

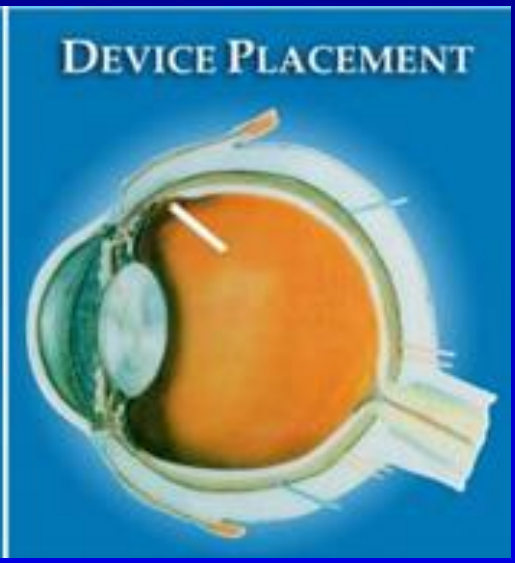
- DHA (docasohexanoic acid)
- Lutein
- ? ß-carotene

- AVOID: Vitamin E, Smoking

Neuroprotection





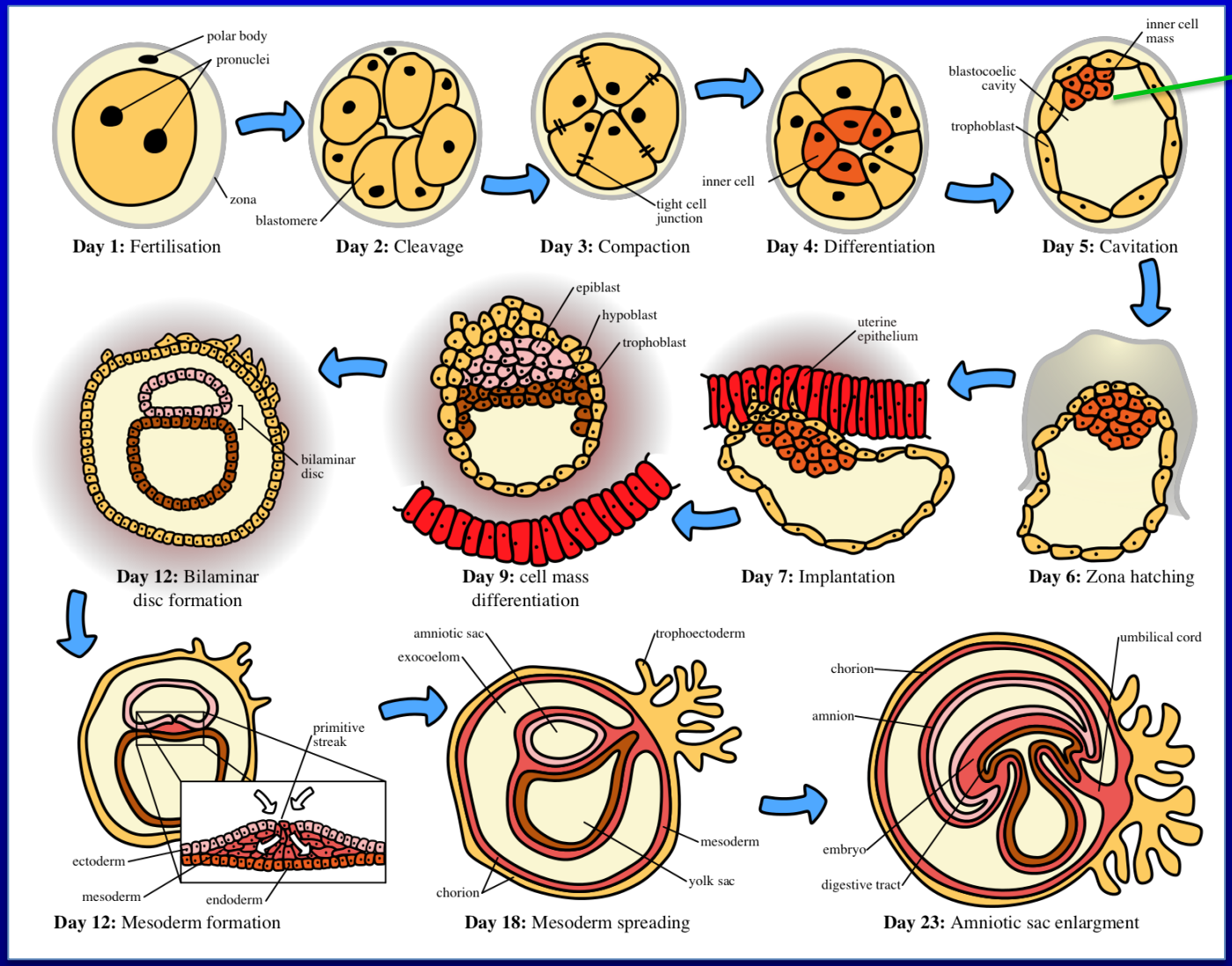


www.medgadget.com

Ciliary Neurotrophic Factor (CNTF)

Results of clinical trials have not been impressive

What is a Stem Cell?



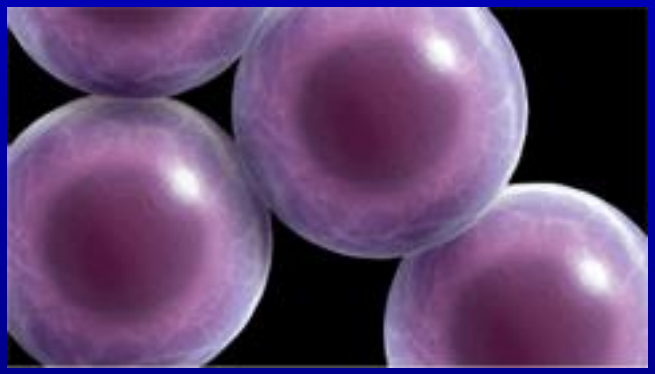
Totipotent

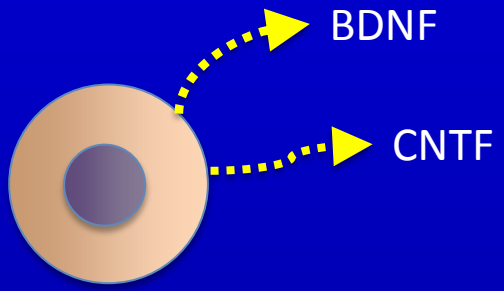
- Unlimited capacity to divide
- Can become any other tissue

Pluripotent

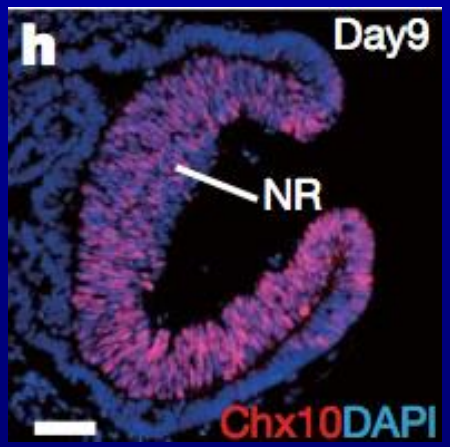
- Unlimited capacity to divide
- Can become a subset of tissues

How stem cells might work…





Release Protective Factors



Differentiate in Retinal Tissue

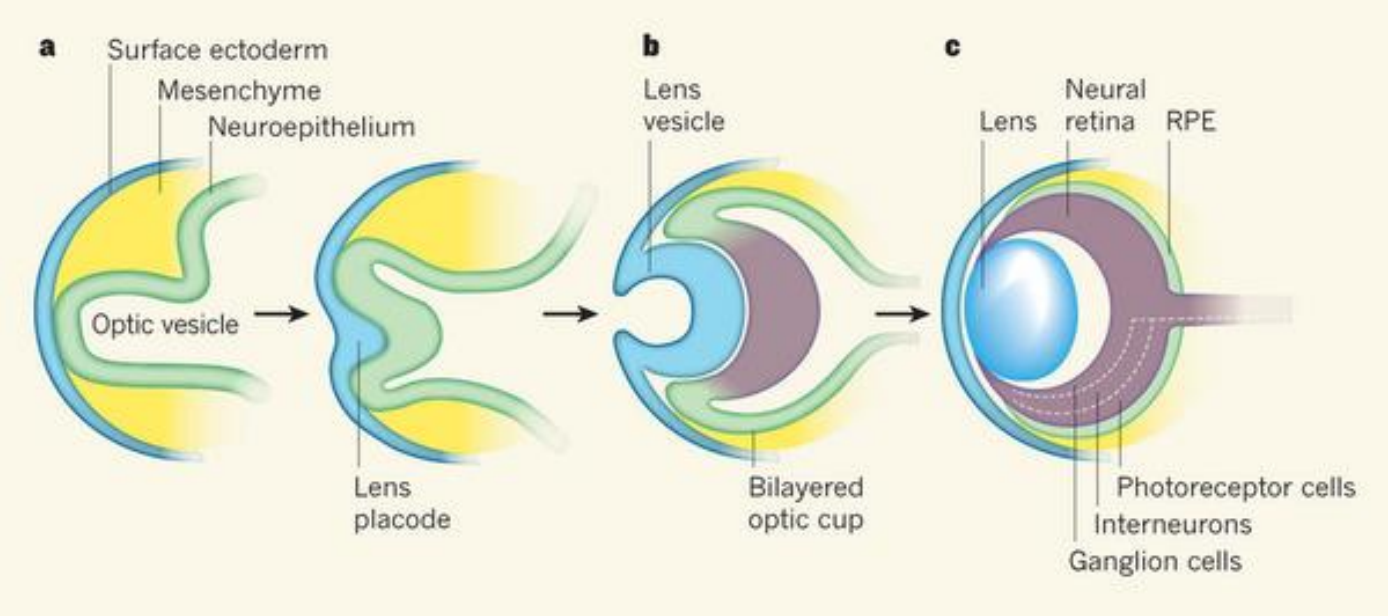
Image from Eriraku et al. 2011

Current Stem Cell Trials

Sponsor	Cell Derivation	Differentiation	Delivery	Disease	Sites
ACT	Embyronic	RPE	subretinal	Stargardt	UCLA, Bascom, Wills, MEEI, Moorfields, Korea
ACT	Embyronic	RPE	subretinal	AMD	UCLA, Bascom, Wills, MEEI, Korea
Pfizer	Embyronic	RPE	subretinal scaffold	AMD	UCL
Stem Cells Inc.	CNS		subretinal	AMD	RFSW
	Bone Marrow		intravitreal	RP, AMD	Univ. Sao Paulo
	Bone Marrow		intravitreal	RP	Madihol Univ (Thailand)
	Bone Marrow	CD34+	intravitreal	RP, DM, AMD	UC Davis

Source: www.clinicaltrials.gov

How to build a new retina



Self-Formation of Optic Cups and Storable Stratified Neural Retina from Human ESCs

Tokushige Nakano,[1,2,4,5] Satoshi Ando,[1,2,4] Nozomu Takata,[1] Masako Kawada,[1] Keiko Muguruma,[1] Kiyotoshi Sekiguchi, Koichi Saito,[4] Shigenobu Yonemura,[3] Mototsugu Eiraku,[1,2] and Yoshiki Sasai[1,2,5,*]

[1]Organogenesis and Neurogenesis Group
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RIKEN Center for Developmental Biology, Kobe 650-0047, Japan
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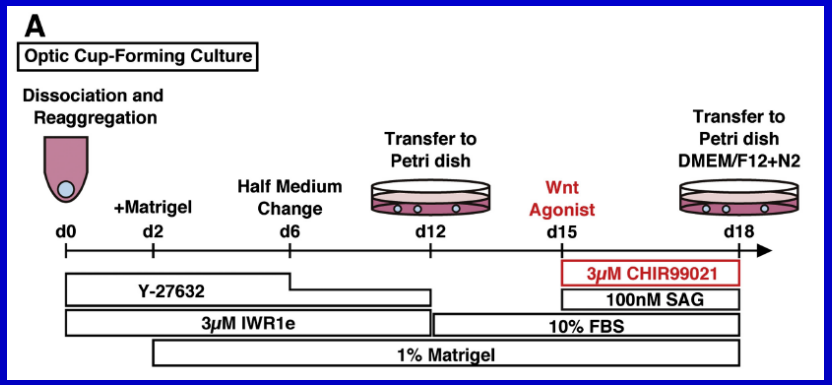
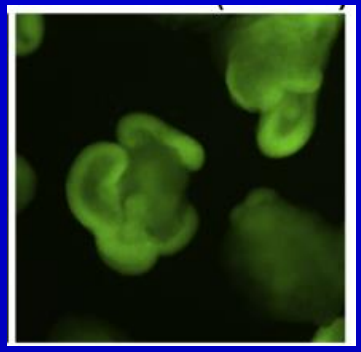
[6]Laboratory of Extracellular Matrix Biochemistry, Institute for Protein Research, Osaka University, Suita 565-0871, Japan
*Correspondence: yoshikisasai@cdb.riken.jp

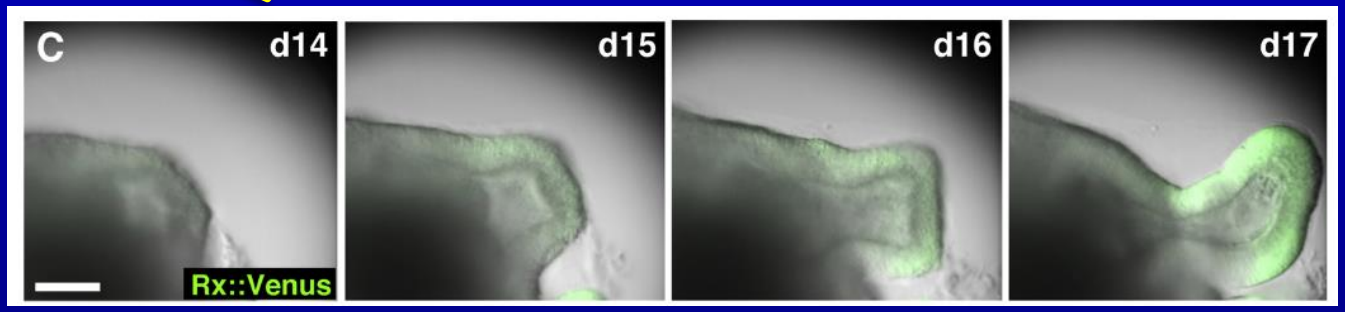


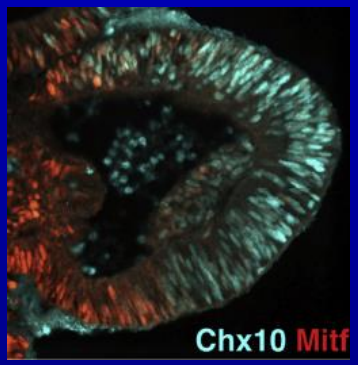
hESC grown in 3D Matrigel with SFEBq media

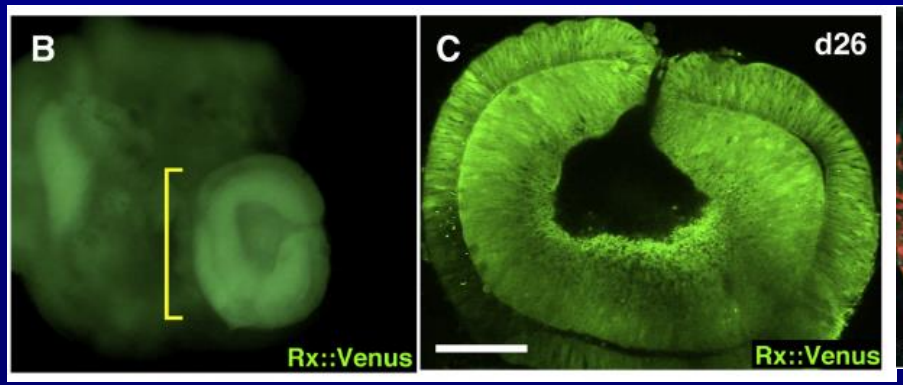
Transfect venus under RX promoter

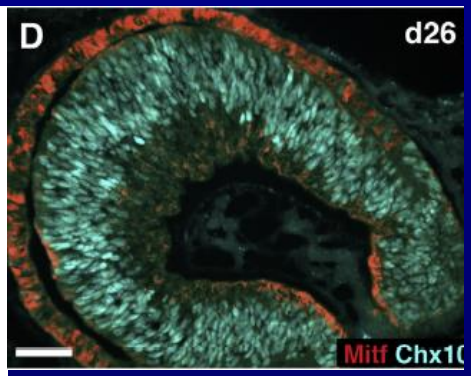


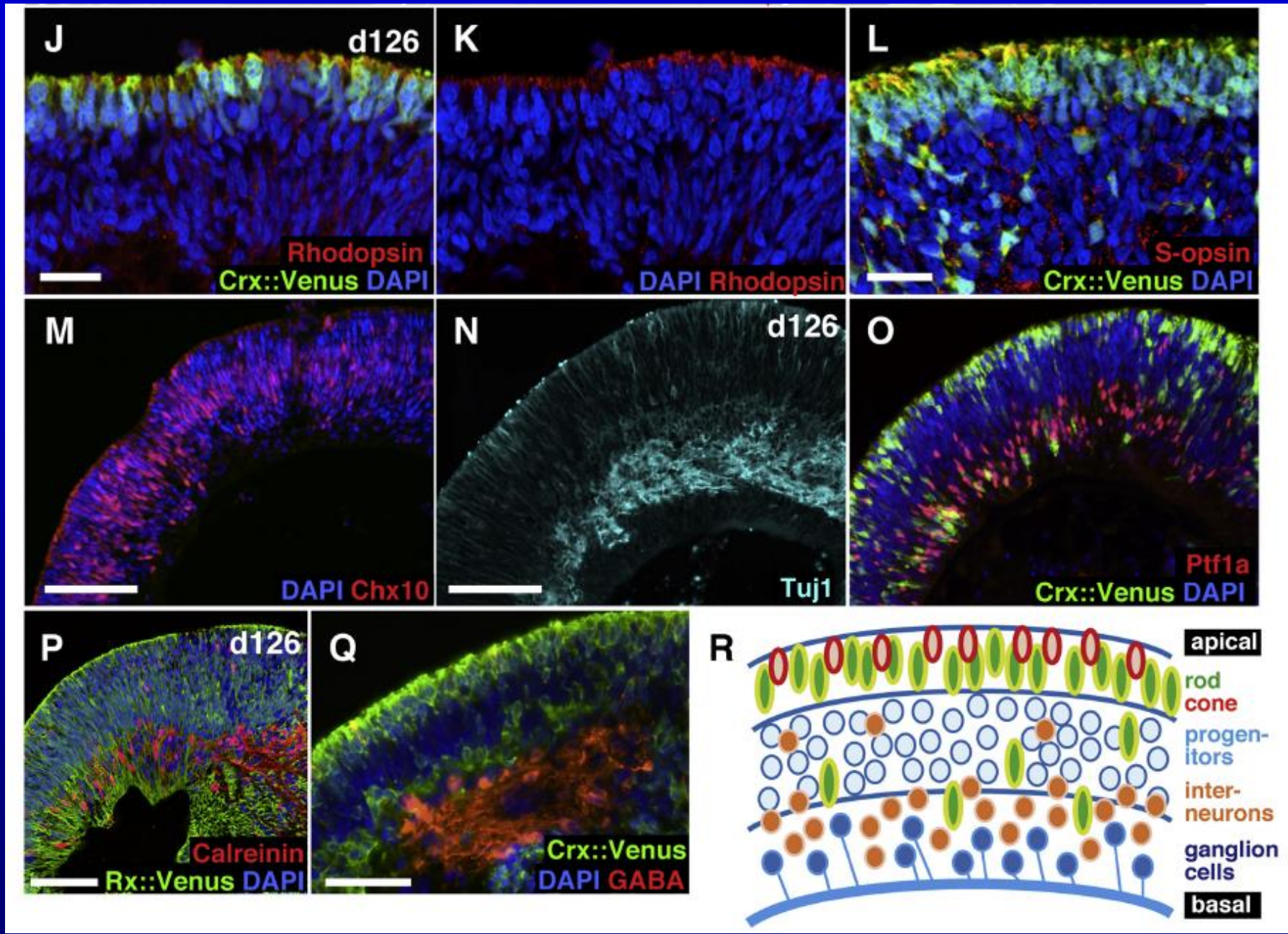
FACS for RX:Venus positive cells










J
d126
Rhodopsin
Crx::Venus DAPI
K
DAPI Rhodopsin
L
S-opsin
Crx::Venus DAPI
M
DAPI Chx10
N
d126
Tuj1
O
Ptf1a
Crx::Venus DAPI
P
d126
Calreinin
Rx::Venus DAPI
Q
Crx::Venus
DAPI GABA
R
apical
rod
cone
progen-
itors
inter-
neurons
ganglion
cells
basal

A Novel Approach for Subretinal Implantation of Ultrathin Substrates Containing Stem Cell-Derived Retinal Pigment Epithelium Monolayer

Yuntao Hu[a, e] Laura Liu[a, f] Bo Lu[c] Danhong Zhu[a, b] Ramiro Ribeiro[a, g]
Bruno Diniz[a, h] Padmaja B. Thomas[a] Ashish K. Ahuja[a] David R. Hinton[a, b]
Yu-Chong Tai[c] Sherry T. Hikita[d] Lincoln V. Johnson[d] Dennis O. Clegg[d]
Biju B. Thomas[a] Mark S. Humayun[a]

Ophthalmic Res 2012;48:186–191

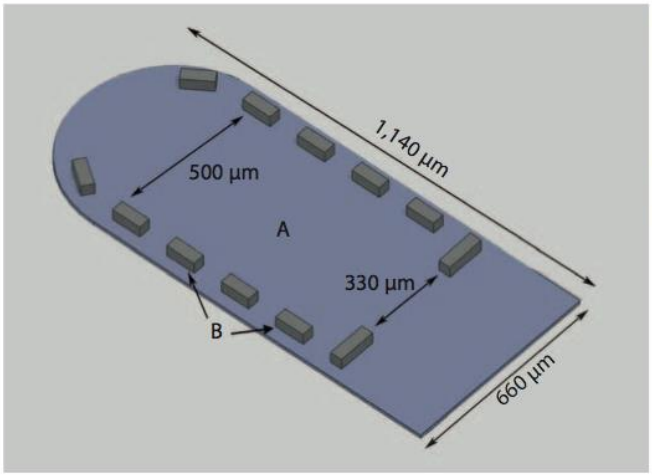


Fig. 1. Diagrammatic sketch of the implantation tool. The device consists of a thin parylene plate (A) containing barriers (B) arranged in the form of a 'U'. The substrate for implantation is placed in the middle of the U-shaped area (substrate chamber).

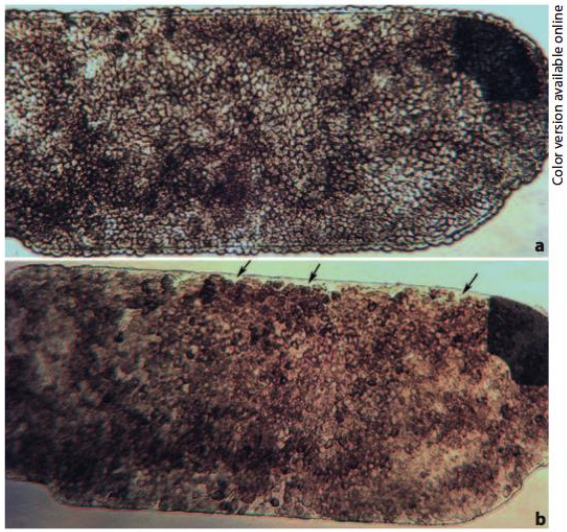
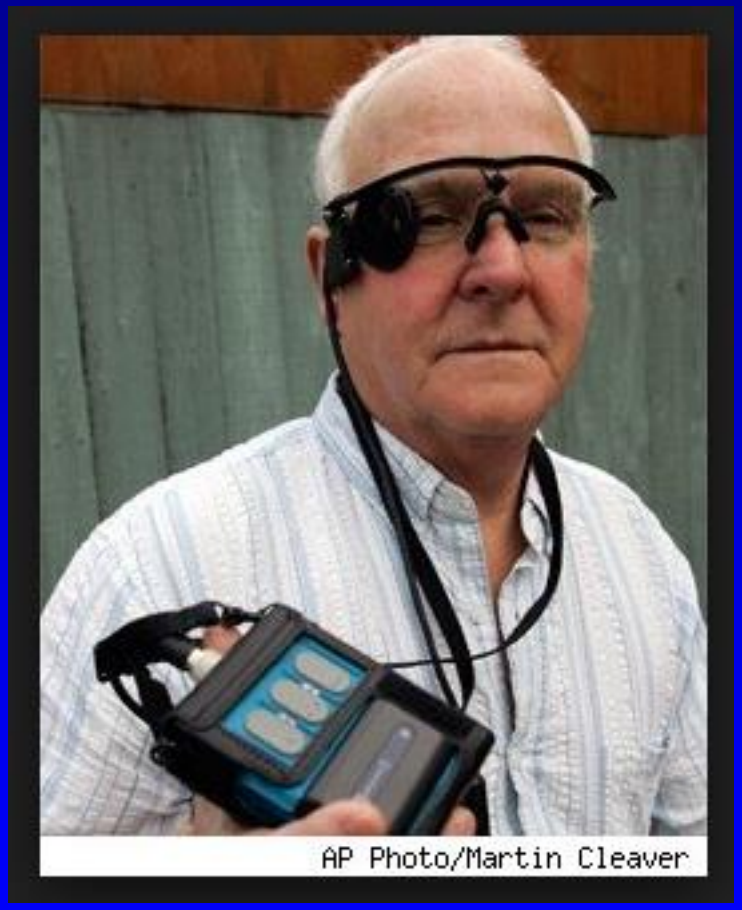


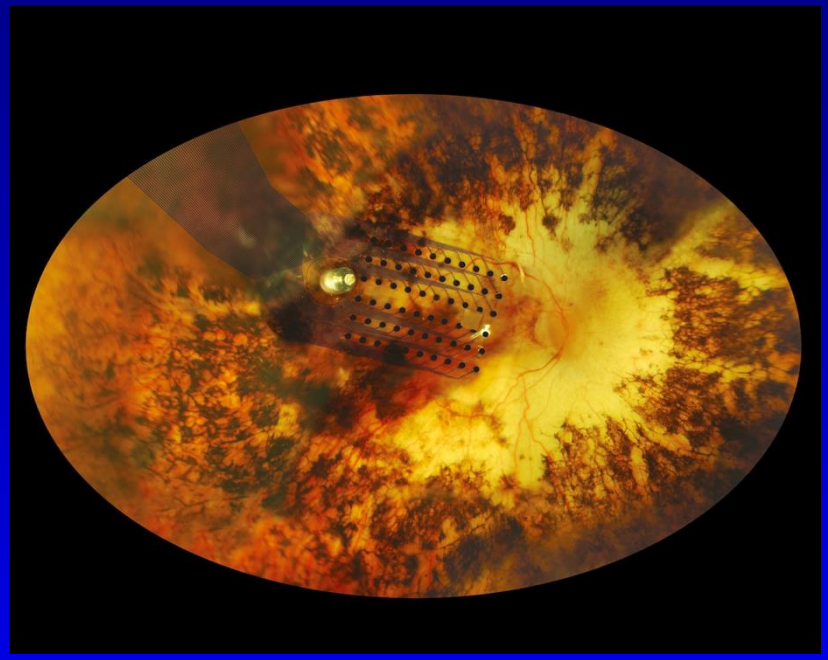
Fig. 2. The ultrathin substrate containing hESC-RPE cells. Images were taken before (**a**) and after (**b**) subretinal implantation. **b** Considerable cell loss can be observed along the edges of the substrate (arrows).

4-µm-thick parylene substrates containing a monolayer of human embryonic stem cell-derived RPE

Implantable Retinal Silicon Chips



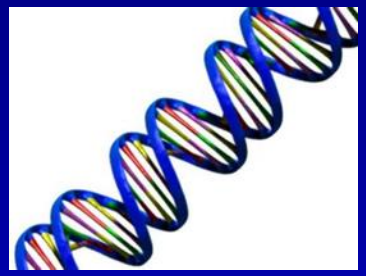
Argus II Retinal Implant



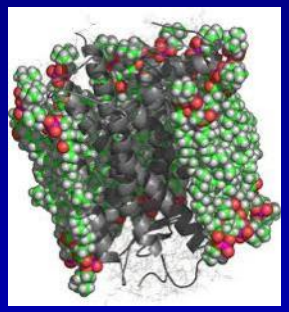
Gene Therapy

What is gene therapy?

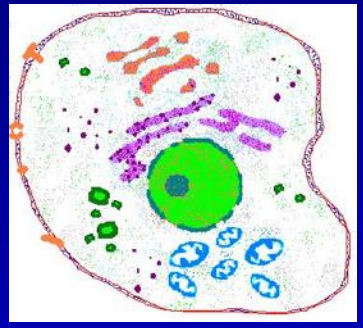
Normal



DNA

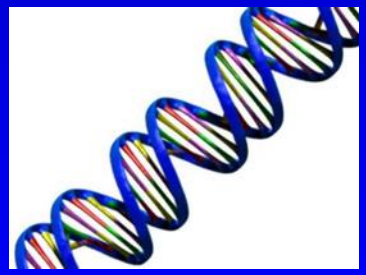


Protein



Cells

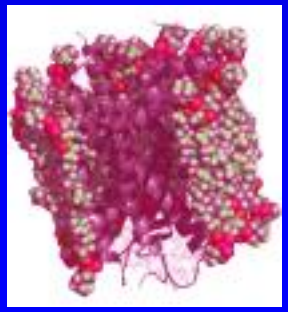
Abnormal



DNA

Mutation

No Protein



Mutant Protein

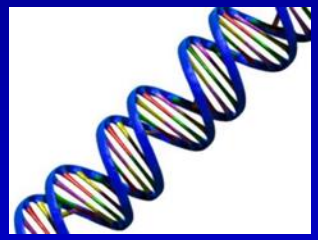
Gene Therapy Approach

Replace missing protein

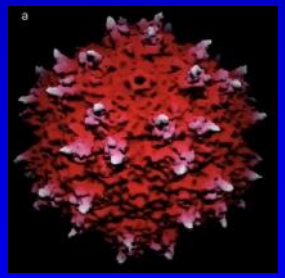
Decrease bad protein

How to Deliver Gene Therapy?

- **Vector** – a mechanism to deliver DNA to a cell



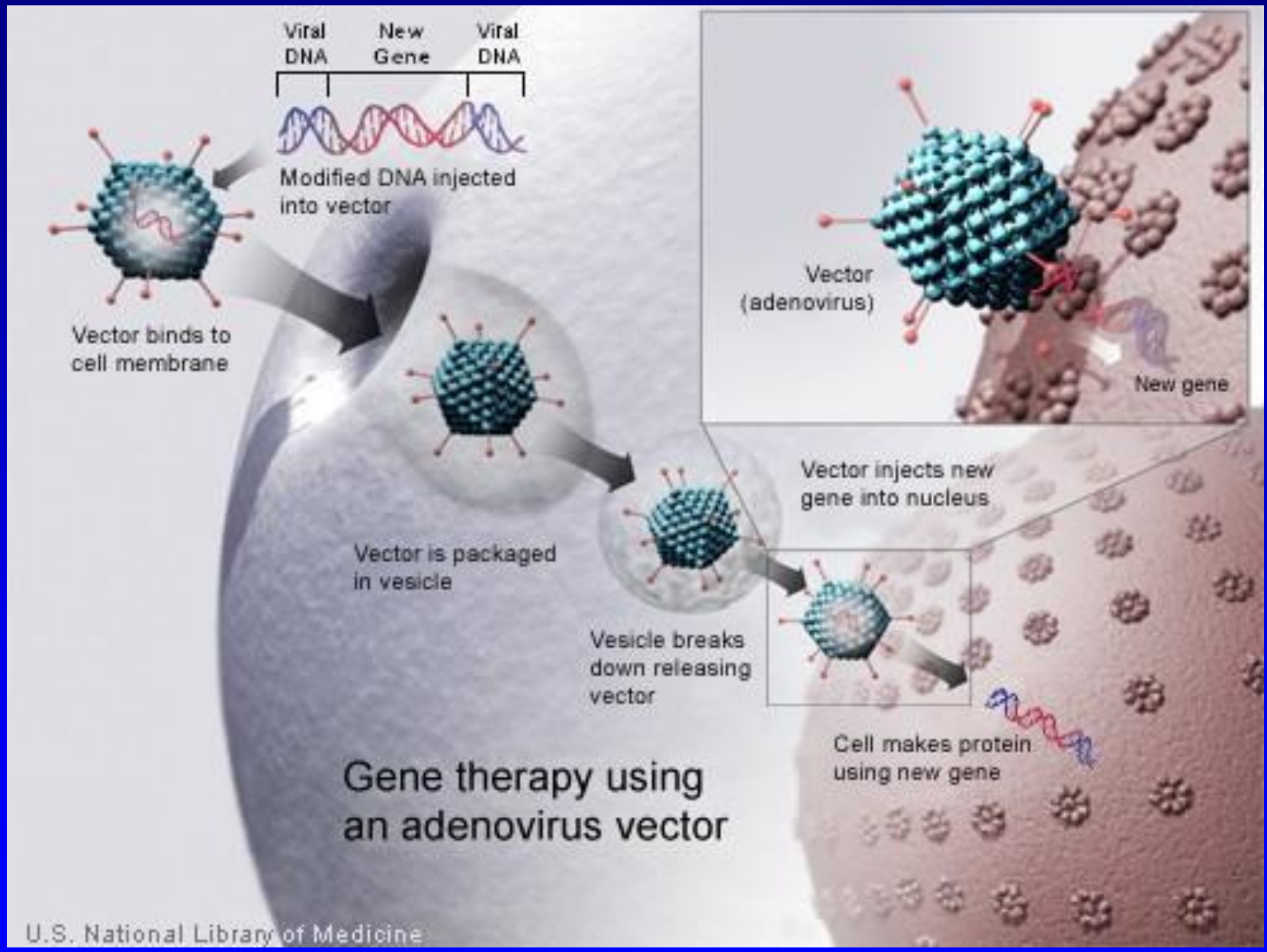
Bare DNA



Modified Virus

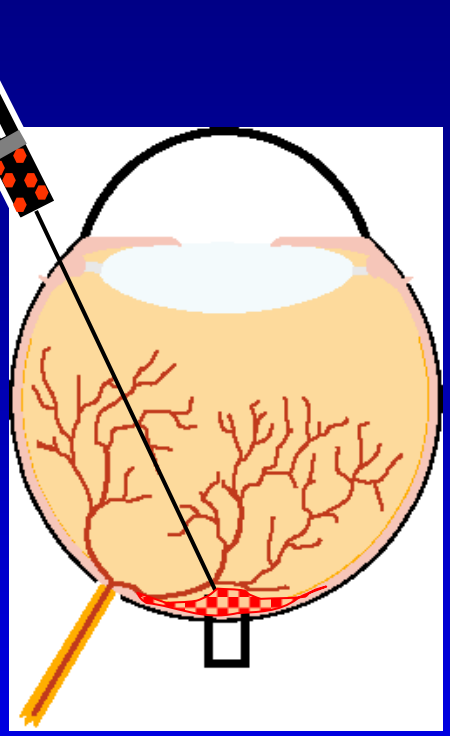
Nanoparticles

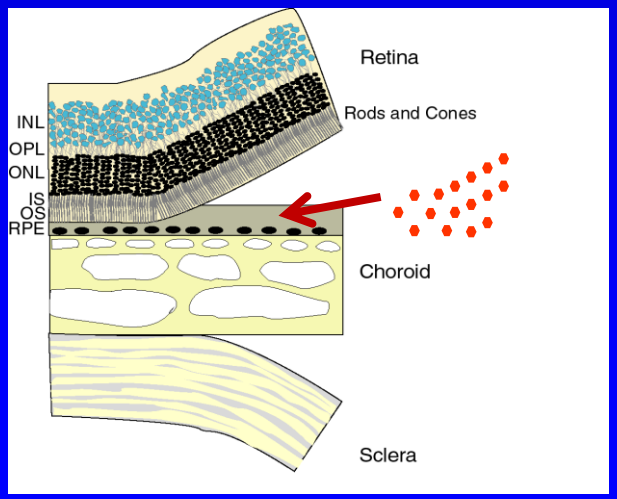
Gene Therapy

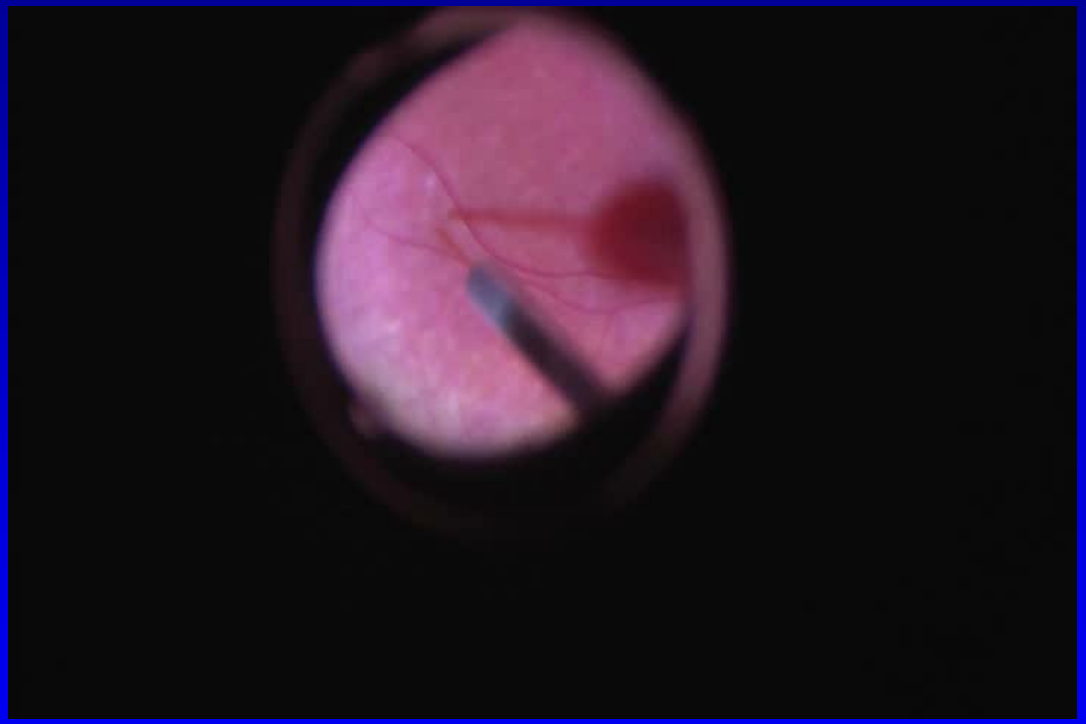


Gene Therapy - Subretinal Injection

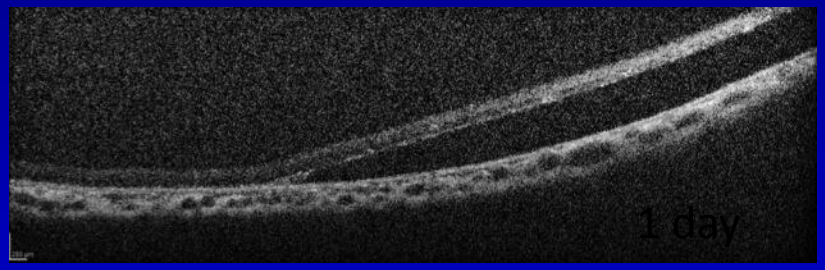
450µL

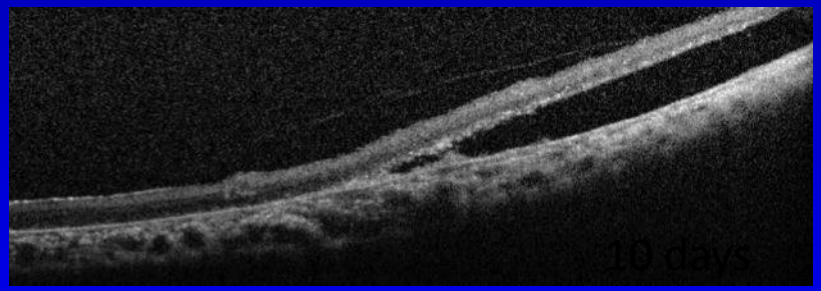


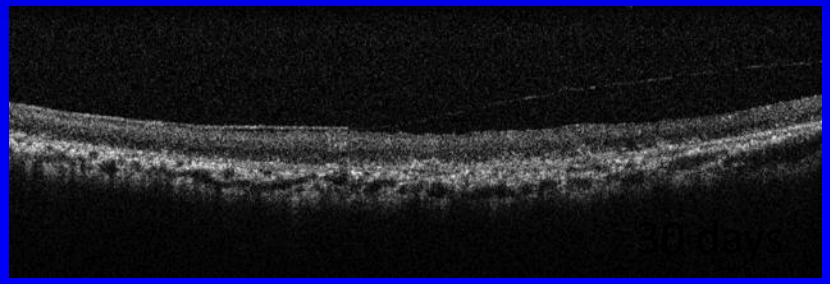




Recovery of Subretinal Bleb







- Immediately after injection
- Bleb resorbed

Gene Therapy Trials at Casey Eye Institute

Current

- Leber Congenital Amaurosis Type 2 (*RPE65*)
- Stargardt Disease (*ABCA4*)
- Usher Syndrome Type 1B (*MYO7A)*
- Retinostat (Endostatin/Angiostatin for NVAMD)

Future/Planned Trials

- X-Linked Retinoschisis (*RS1)*
- Achromatopsia (*CNGB3*)

Gene Therapy Trials at Other Centers

- LCA (RPE65) – University of Pennsylvania, CHOP, Moorfields, Israel
- Choroideremia (*REP1*) – University of Oxford
- Retinitis Pigmentosa (*MERTK*) – King Khalid Eye Specialist Hospital

UshStat

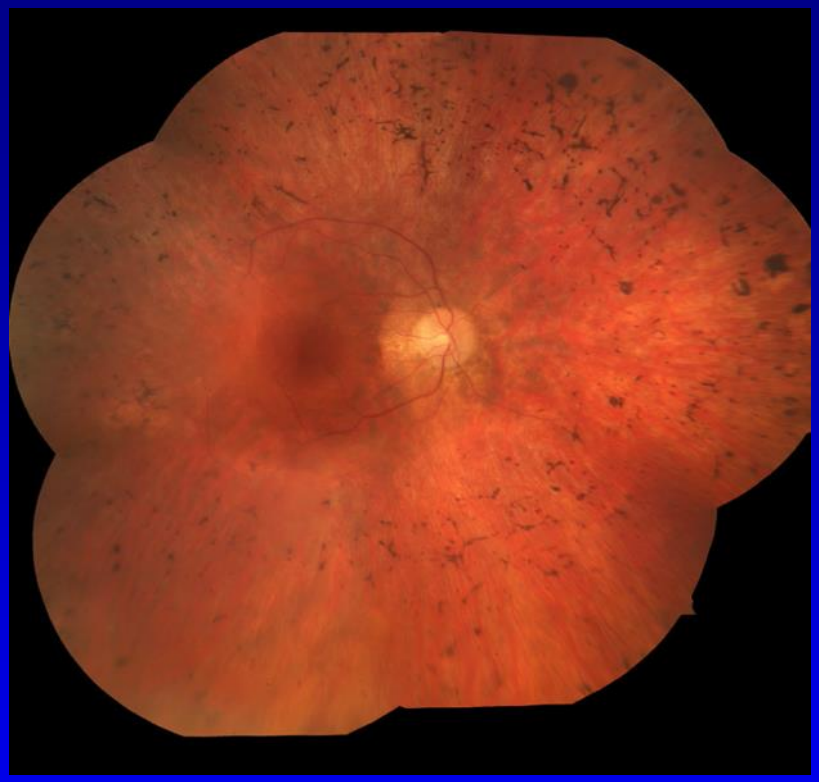
(For Type 1B Usher Syndrome)

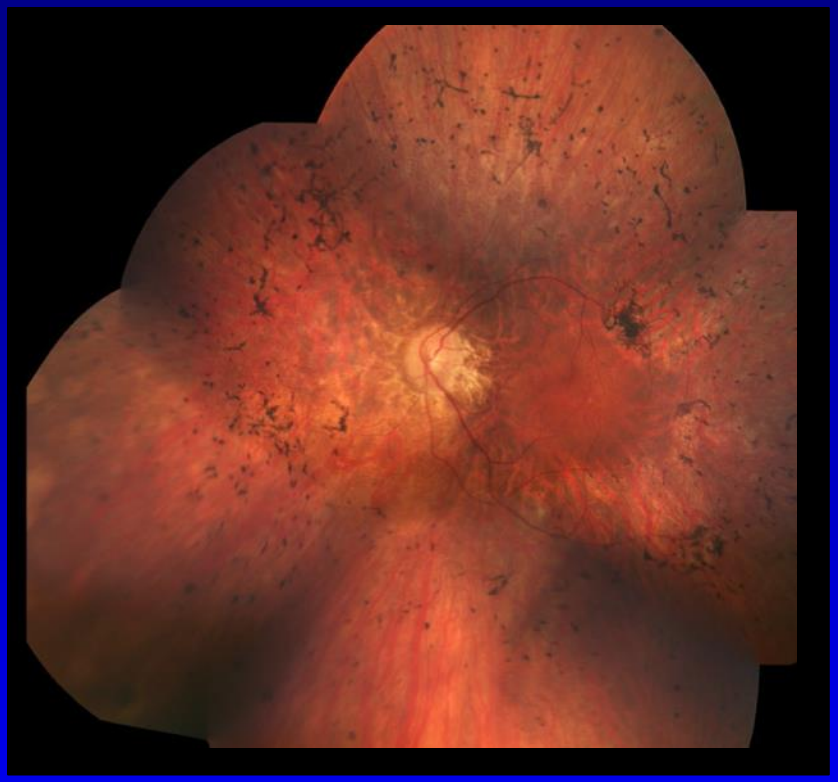
Type 1B Usher Syndrome

Genetic Defect
- Autosomal Recessive
- Mutations in *MYO7A*

Clinical Features
- Severe Early Onset Rod-cone dystrophy
- Severe Congenital Deafness
- Balance problems

Usher Syndrome – Type 1B

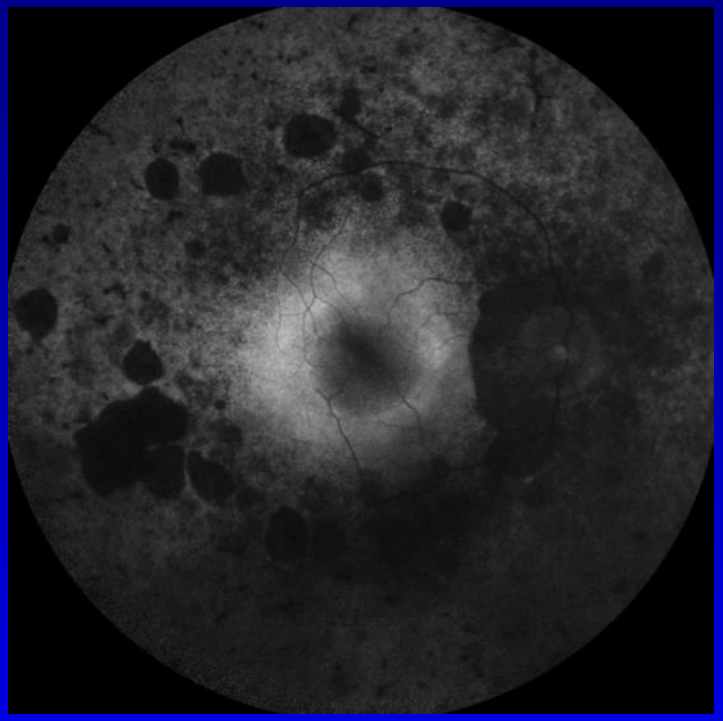


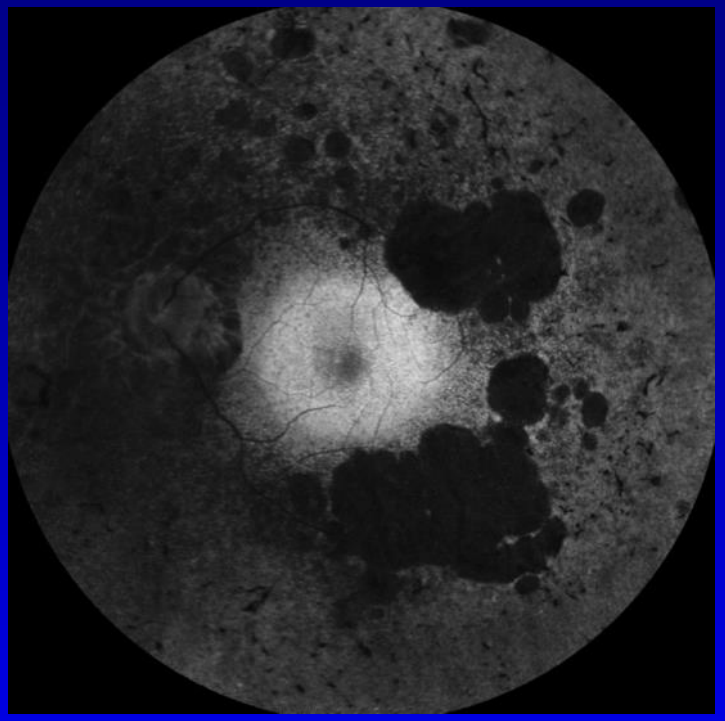


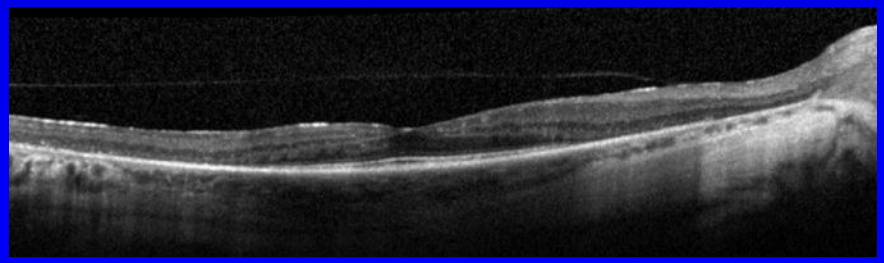
61 yr Female, Va = 20/40 OU

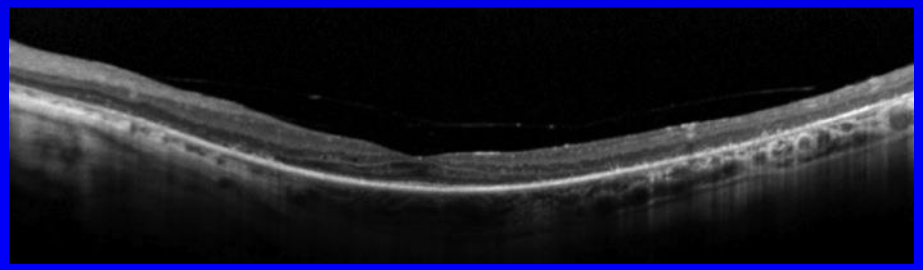
Severe Deafness

Usher Syndrome – Type 1B









61 yr Female, Va = 20/40 OU

A Phase I/IIa Dose Escalation Safety Study of Subretinally Injected USHStat™, Administered to Patients with Usher Syndrome Type 1B

Sponsor: Oxford Biomedica UK / Sanofi

Primary Investigator: Richard Weleber MD

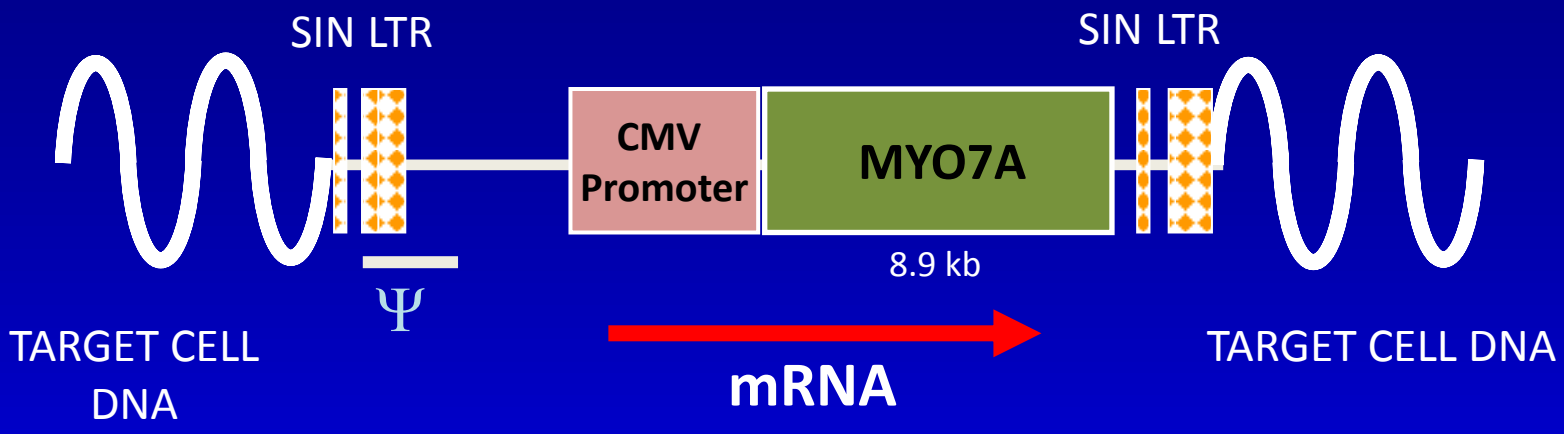
Design: Phase I/IIa dose escalation study

Sites: Casey Eye Institute, OHSU
Hospitalier Nationale d'Ophthalmologie des Quinze-Vingts

Vector: non-primate lentiviral vector based on EIAV

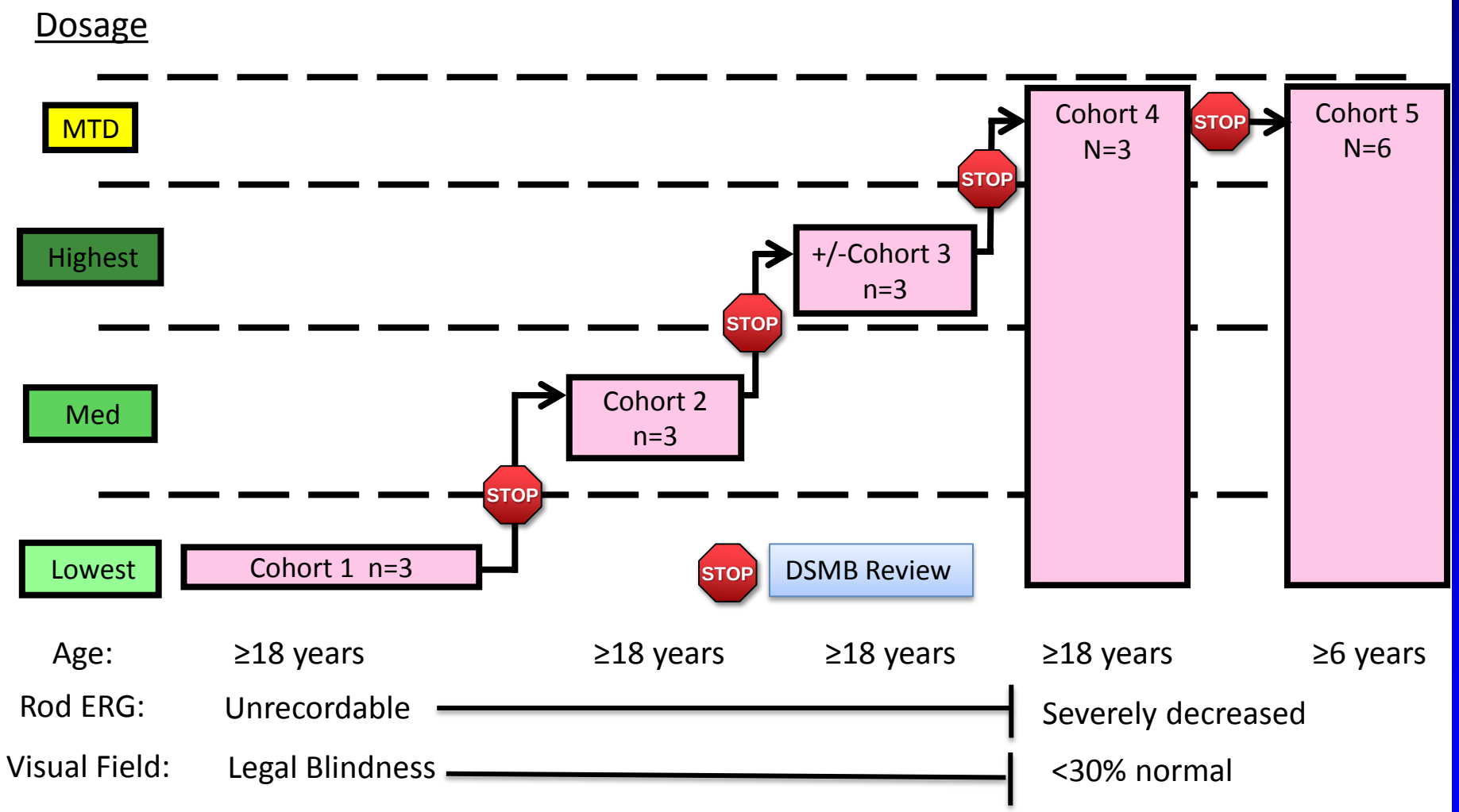
Delivery: Subretinal injection

Ushstat^{tm}



- Based on Equine Infectious Anemia Virus (EIAV)
  - Wild-type virus causes transient anemia in horses, non-pathogenic in humans
  - Ushstat^{tm} vector contains <10% of original viral genome
  - Non-replicating, but does integrate
- Safety/Transfection Studies in Non-human Primates
  - Minimal inflammation
  - Low risk for insertional mutagenesis

Ushstat – Study Design



18 Patients Total

Ushstat Inclusion Criteria

****All Patients:** Two confirmed *MYO7A* mutations

Cohorts 1, 2, 3

- ≥18 years
- Constriction of Kinetic Visual Field that meets criteria for legal blindness
- No detectable rod ERG

Cohort 4

- ≥18 years
- Kinetic Visual Loss - ≥30% reduction sensitivity volume
- Evidence of severe rod/cone dysfunction on ERG

Cohort 5

- ≥6 years
- Kinetic Visual Loss - ≥30% reduction sensitivity volume
- Evidence of severe rod/cone dysfunction on ERG

Ushstat Endpoints

Primary – Safety

- Visual Acuity
- Examination
- Static and Kinetic Visual Field
- OCT
- Laboratory Parameters

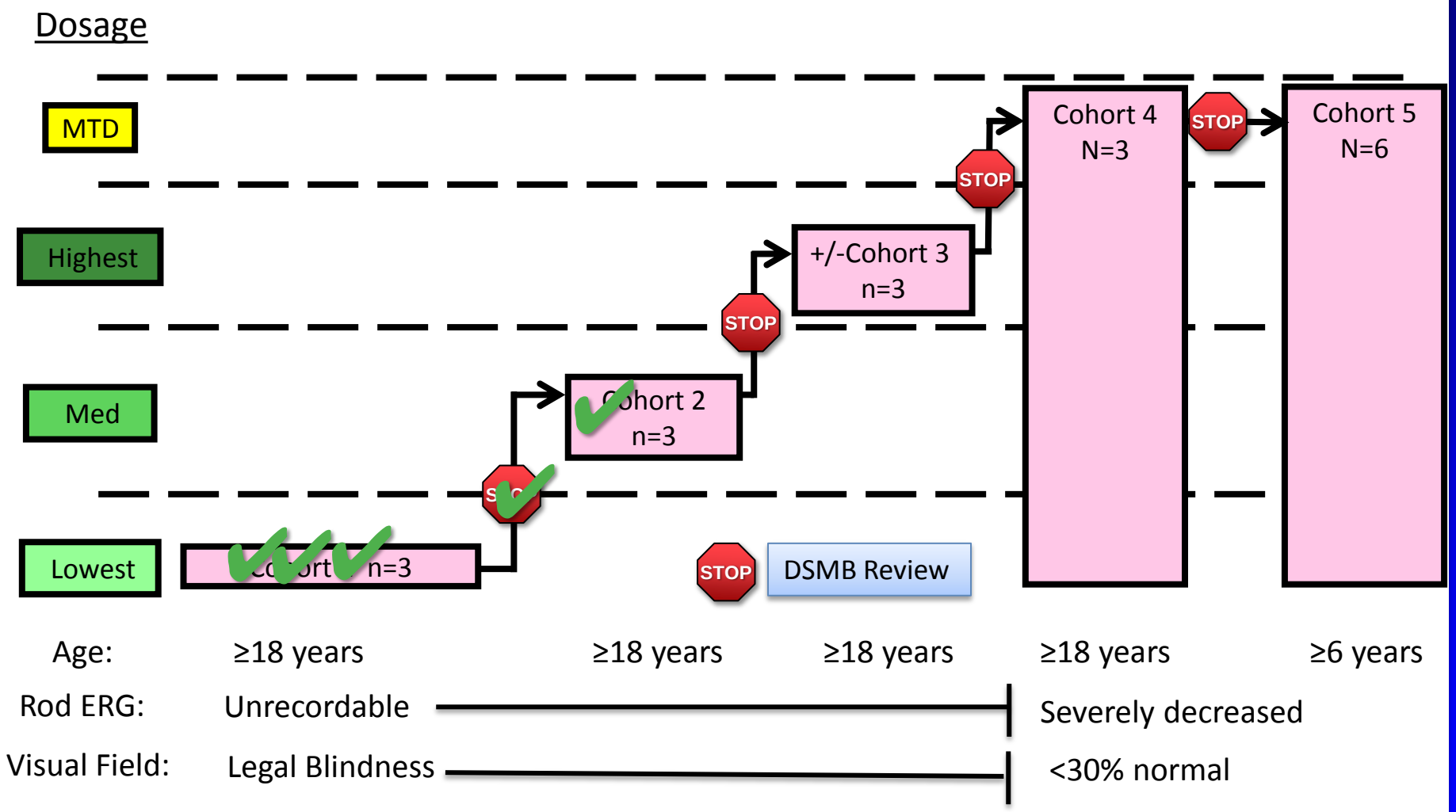
Secondary – Efficacy

- BCVA
- Kinetic and Static Perimetry
- ERG
- OCT
- Adaptive Optics

Recruitment

- First patient dosed April 17, 2012
- Second patient dosed June 28, 2012
- Third patient dosed Oct 4, 2012
- Fourth patient dosed February 16, 2013
- Fifth patient planned, but trial currently on hold

Ushstat – Study Design



- 4 patients treated (3 from Cohort 1, 1 from Cohort 2
- No serious adverse events

Conclusions

- Multiple gene therapy trials are underway with more planned
- Preliminary safety reports are encouraging

Oregon Retinal Degeneration Center
at the Casey Eye Institute

Grant Support

- Hear See Hope
- NIH/NEI 1 K08 EY021186-01
- Foundation Fighting Blindness – CDA
- Research to Prevent Blindness – CDA
- Research to Prevent Blindness
  – Unrestricted grant to CEI
- Foundation Fighting Blindness – Center Grant

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