

# Inherited Progressive Vision Loss

## ABCA4 Responsible Stargardt Disease

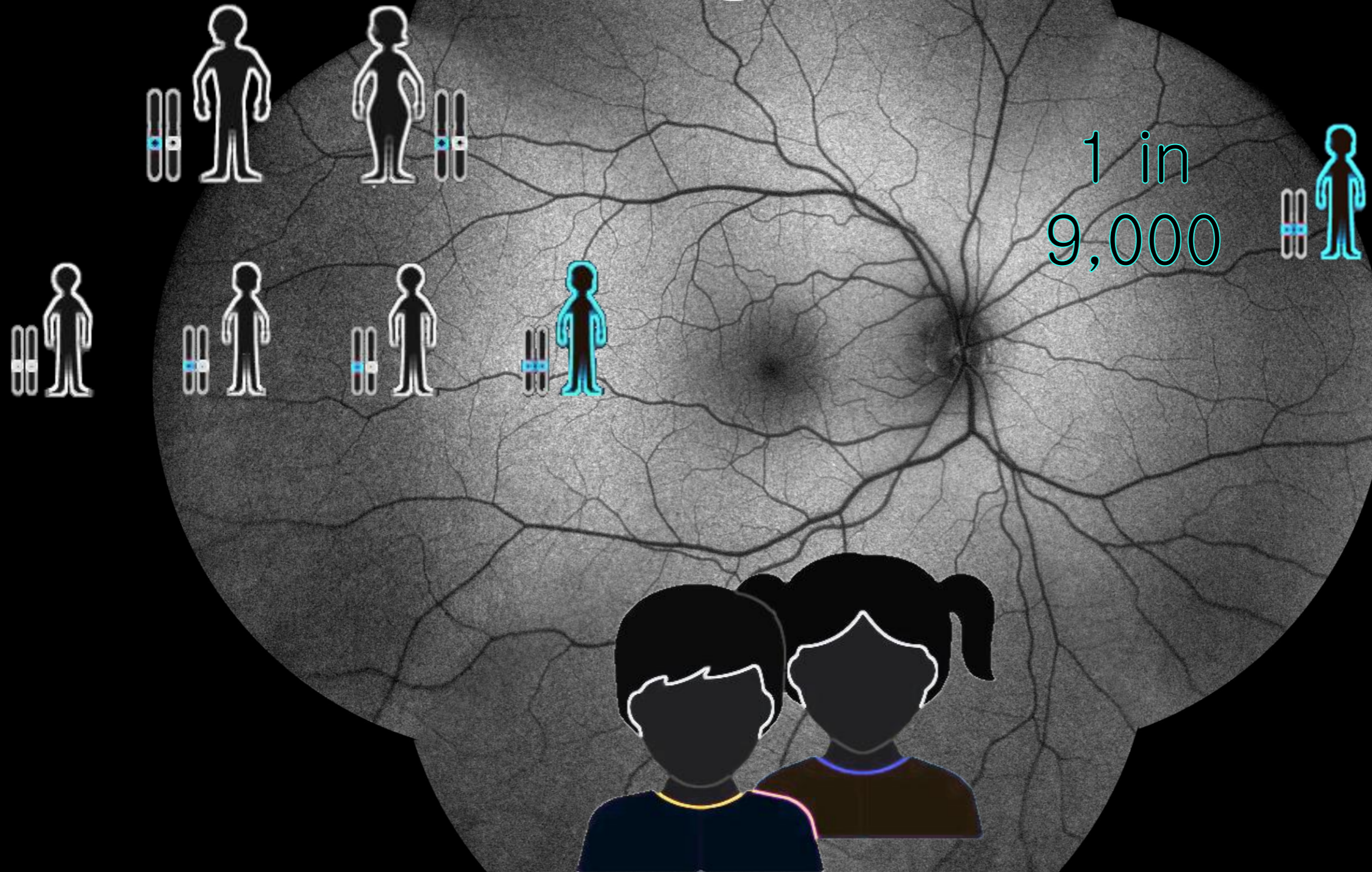


Timothy Biewer-Heisler

Genetics 564 Student  
Project



# What is Stargardt disease?



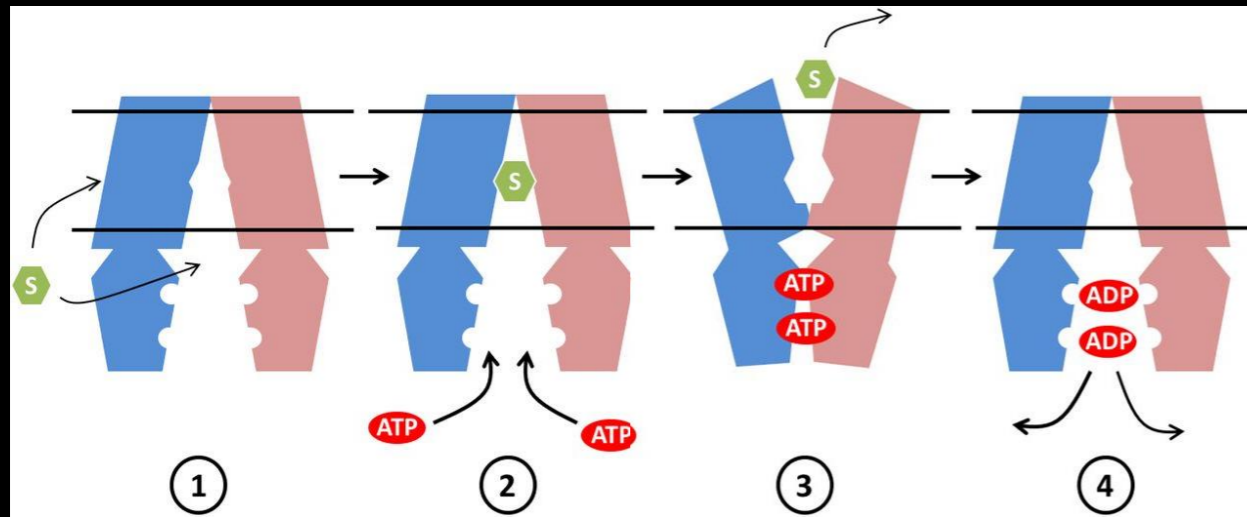
# What are the symptoms of Stargardt disease?

Macular degeneration is the leading cause of impaired reading or detailed vision in the United States. The disease is caused by the breakdown of the central part of the retina. Although macular degeneration affects central and close-up vision, side vision is usually unaffected. Macular degeneration is most common among people over the age of 50. With time, the macula becomes smaller and thinner. This decreases the ability of the macula to function. The macula is a small area of the retina which is responsible for central vision and color vision. Surrounding the macula is the peripheral retina which is responsible for side vision and night vision. Occasionally, macular degeneration is caused by infection or inflammation. The disease may also

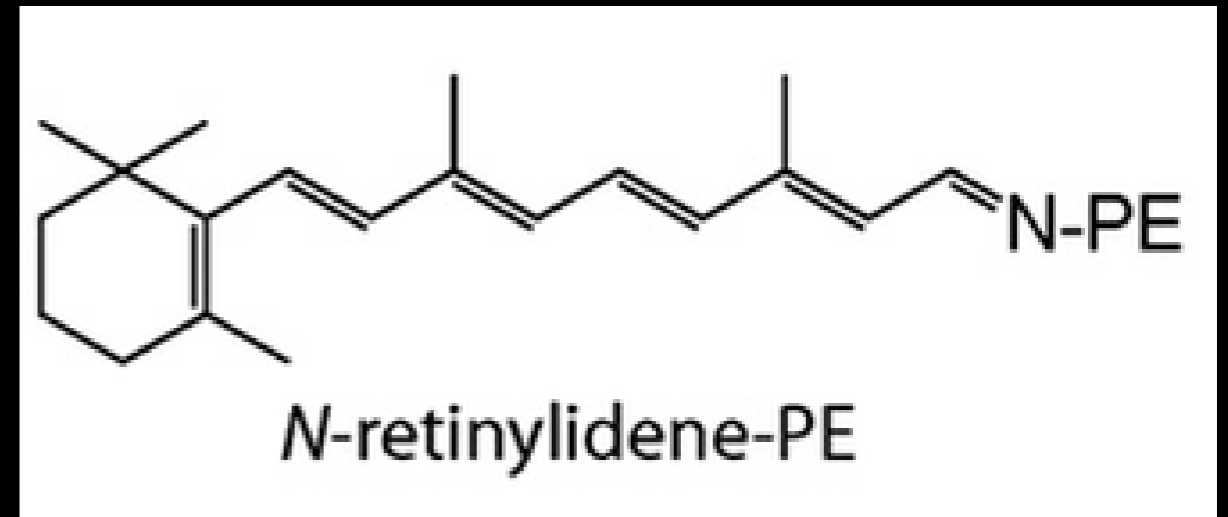


**Lipofuscin buildup**

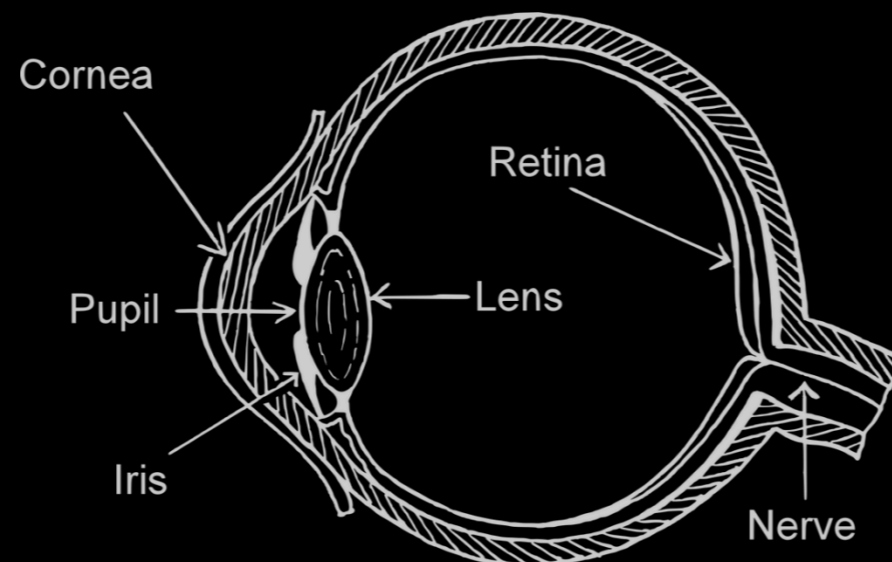
# ABCA4 is responsible for Stargardt disease



**ATP-Binding Cassette**  
**Molecular function**

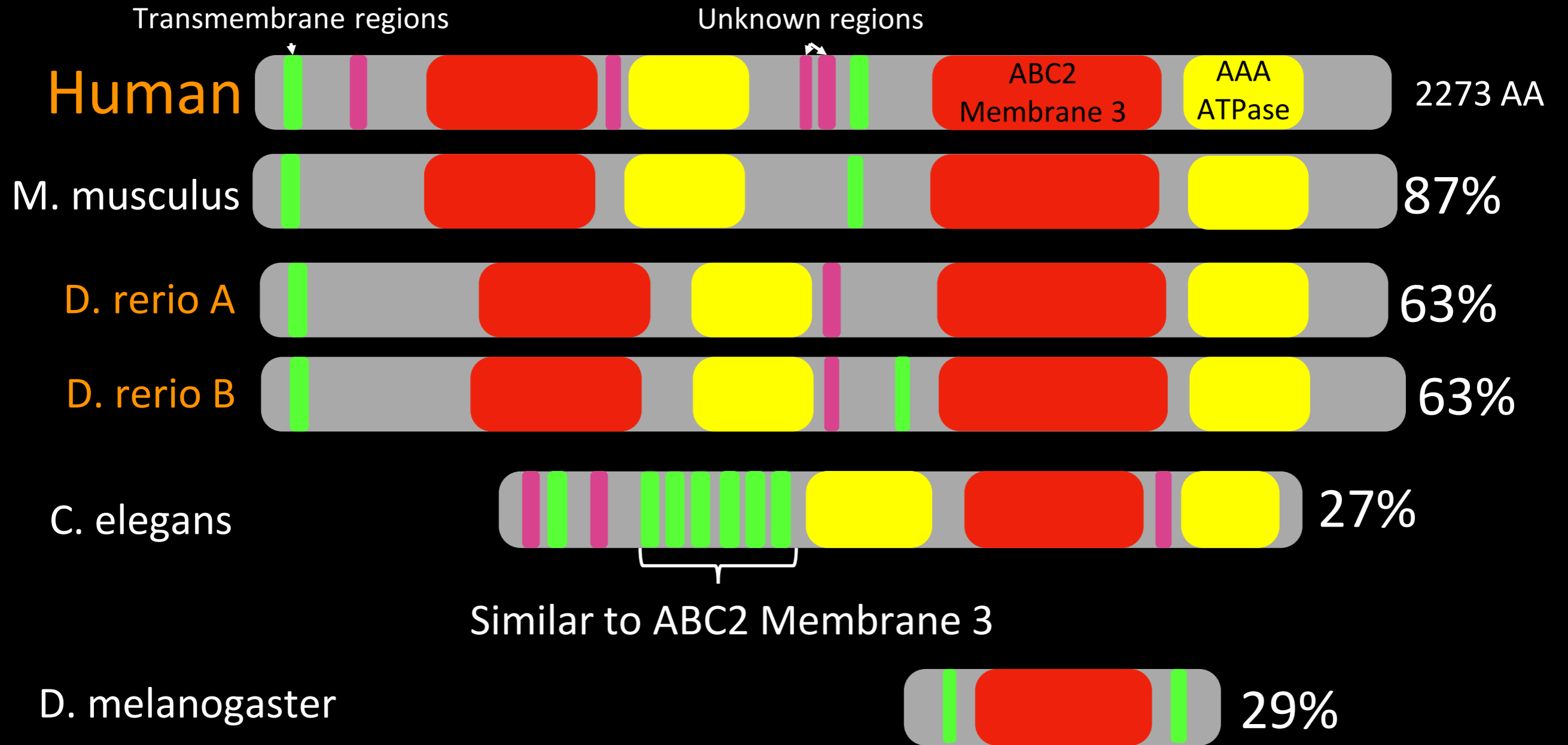


**Lipid Transport on membrane**  
**Cellular component**

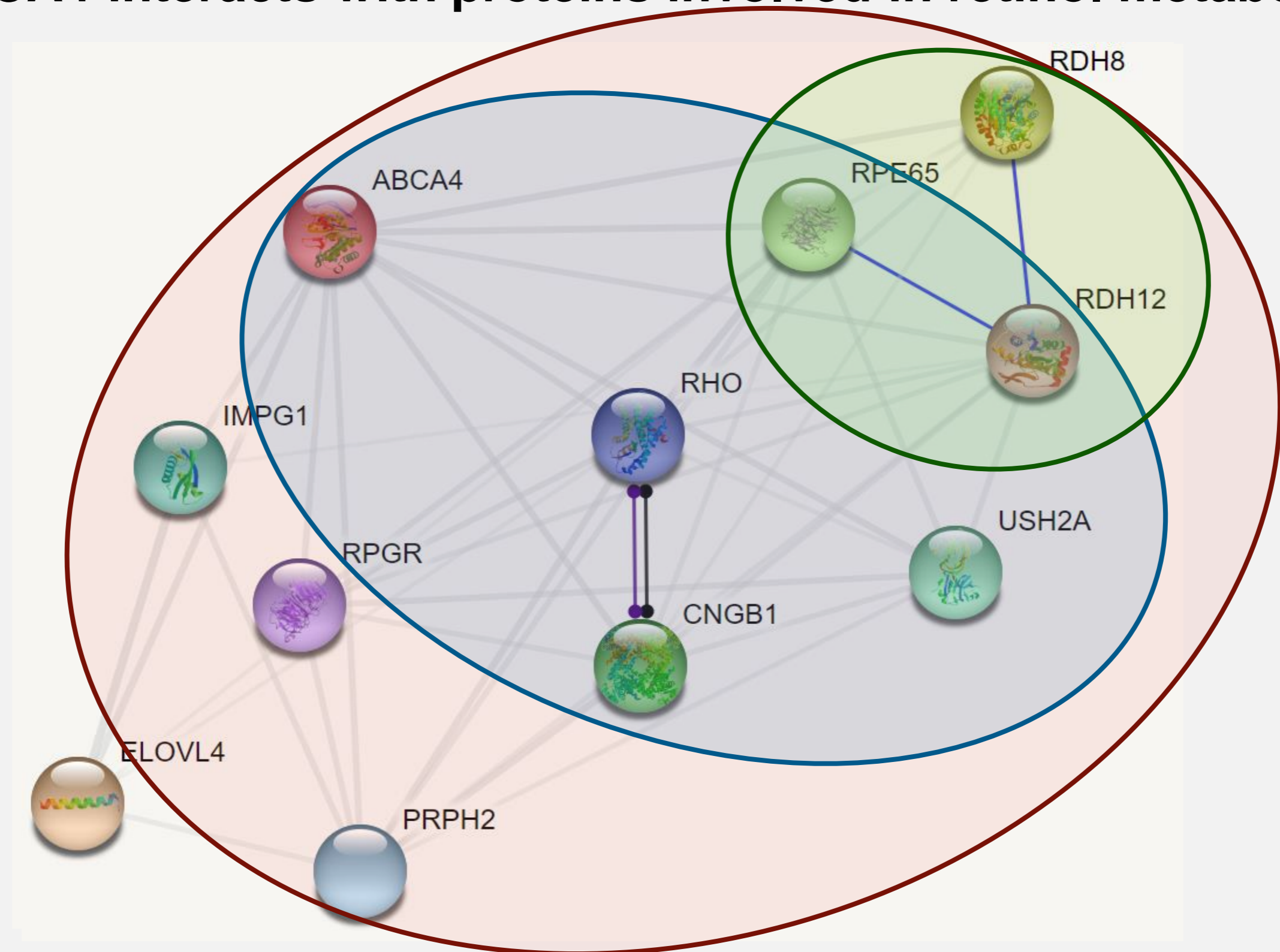


**Visual Perception**  
**Biological process**

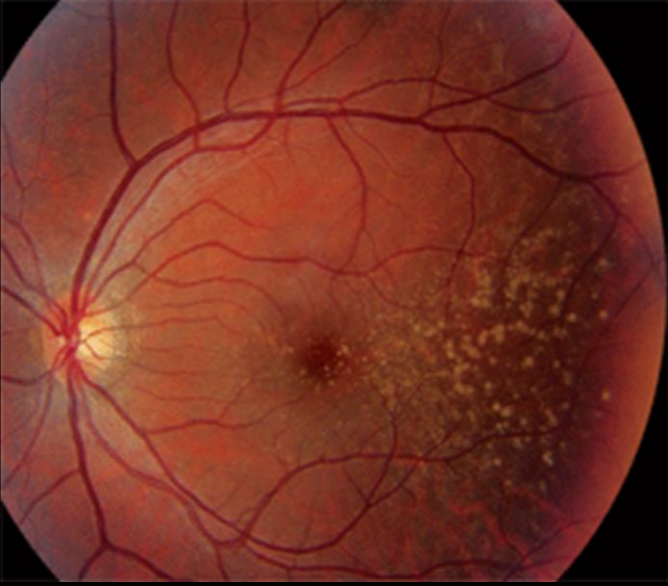
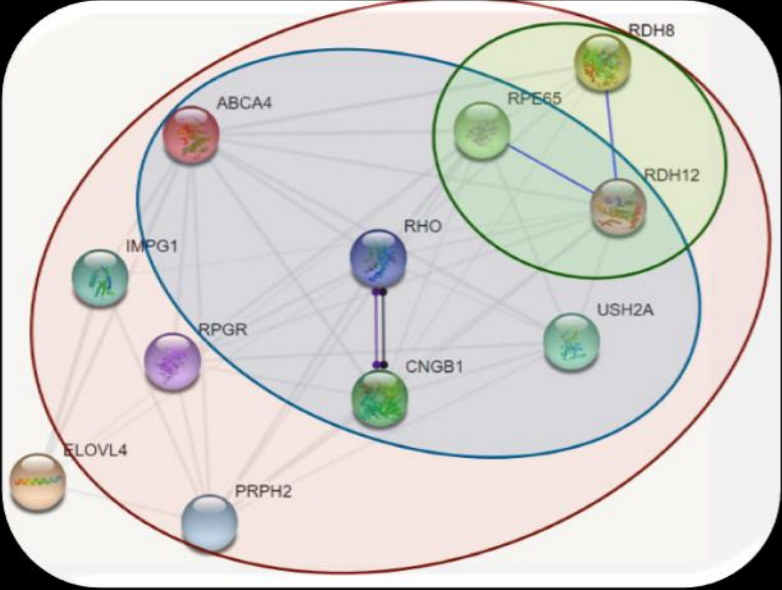
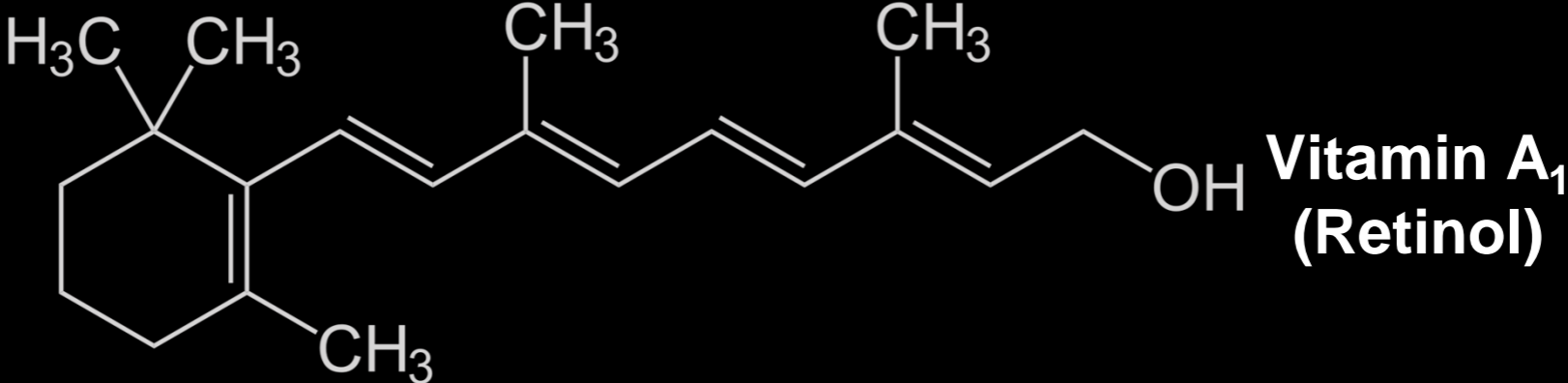
# ABCA4 is conserved between species with...



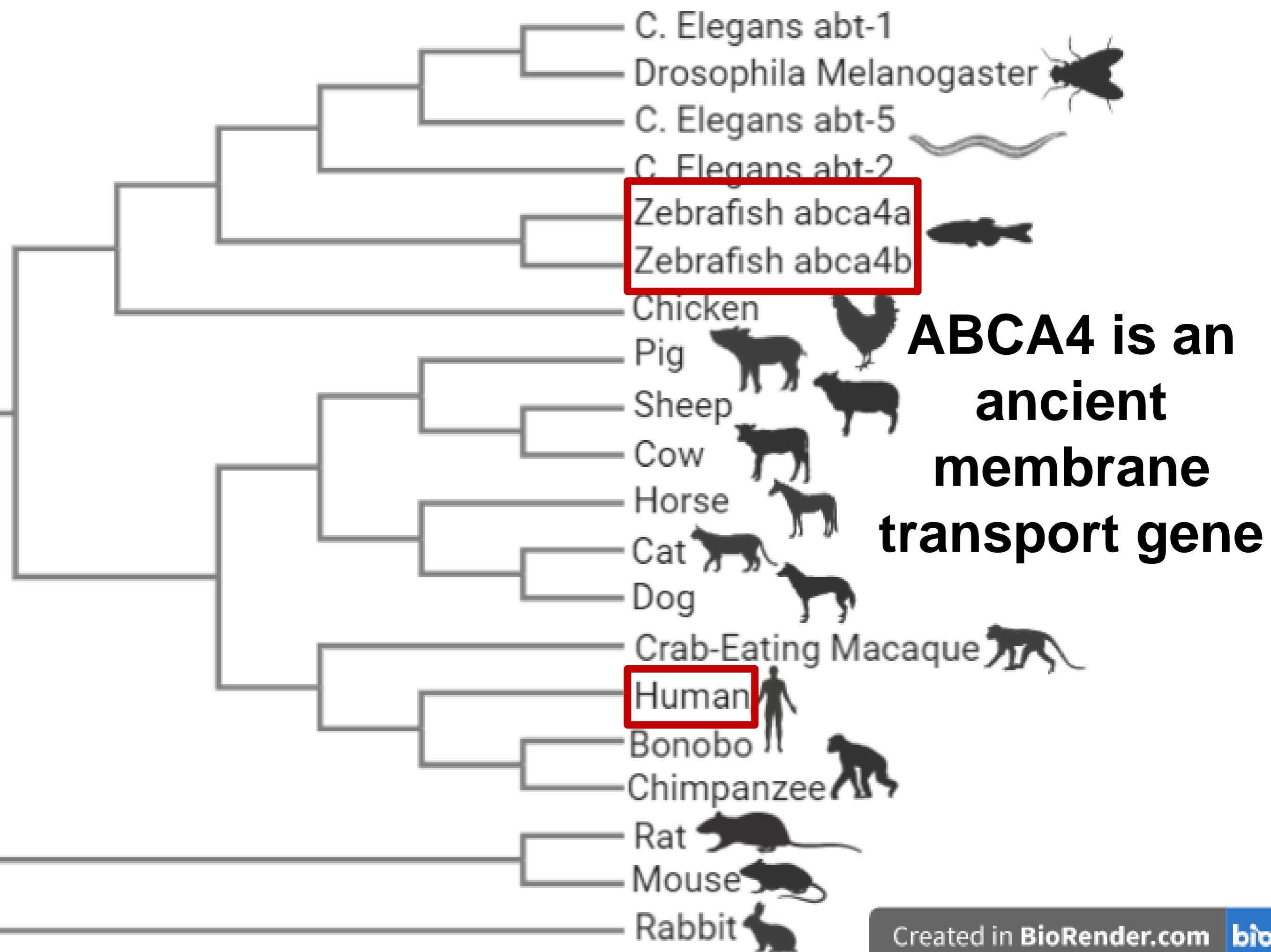
# ABCA4 interacts with proteins involved in retinol metabolism



# Gap: role of ABCA4 in retinol metabolism is unclear

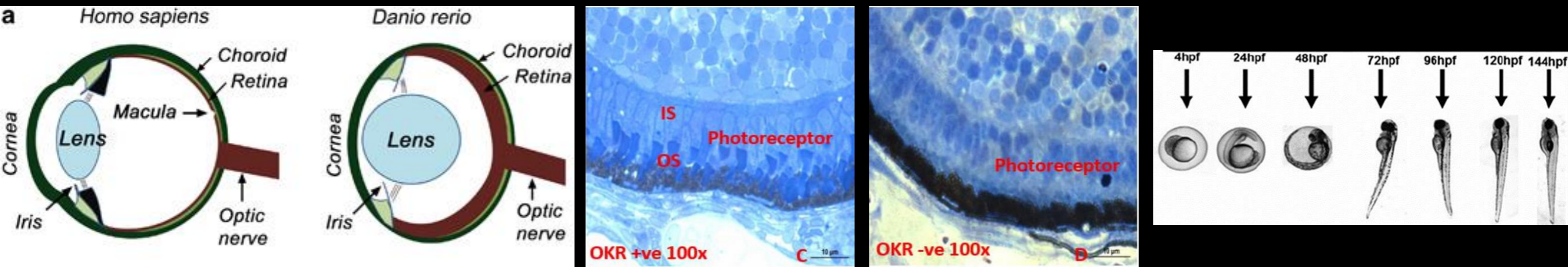


ABCA4-

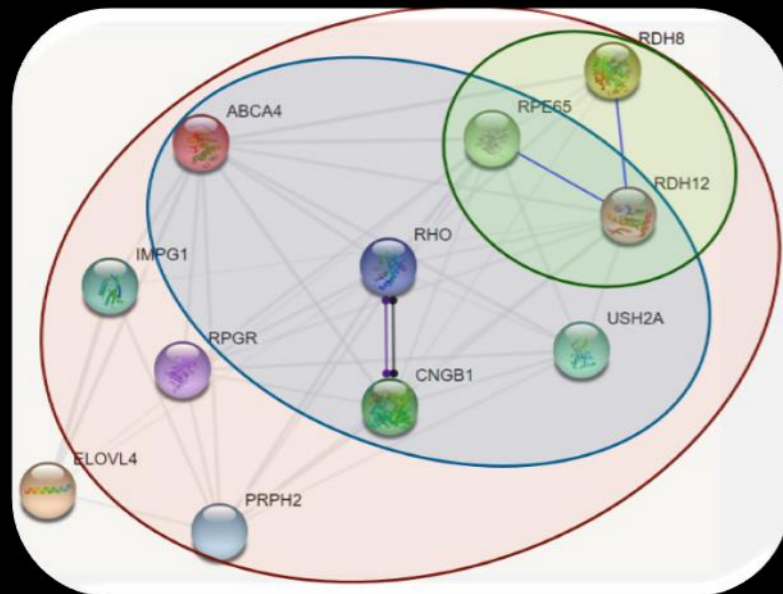
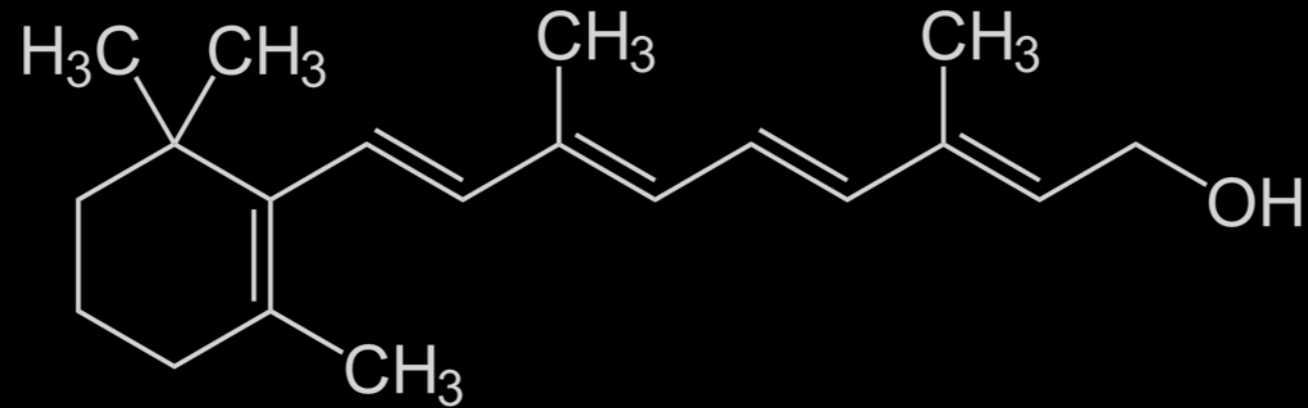




# Zebrafish are excellent models for studying retinol metabolism



# Primary goal: Determine function of ABCA4 in retinol regulation during development



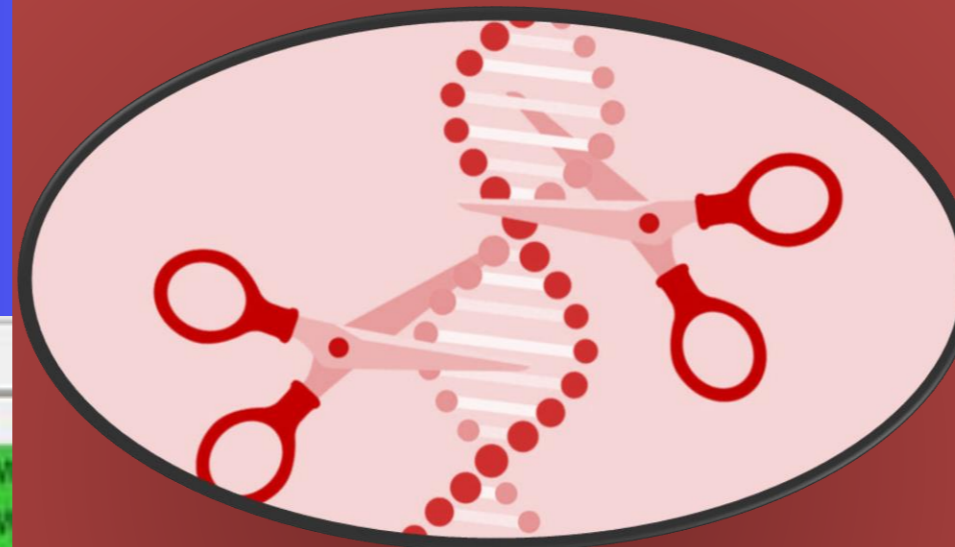
# Aim 1: Determine domains important for retinol metabolism throughout development

## Align sequences

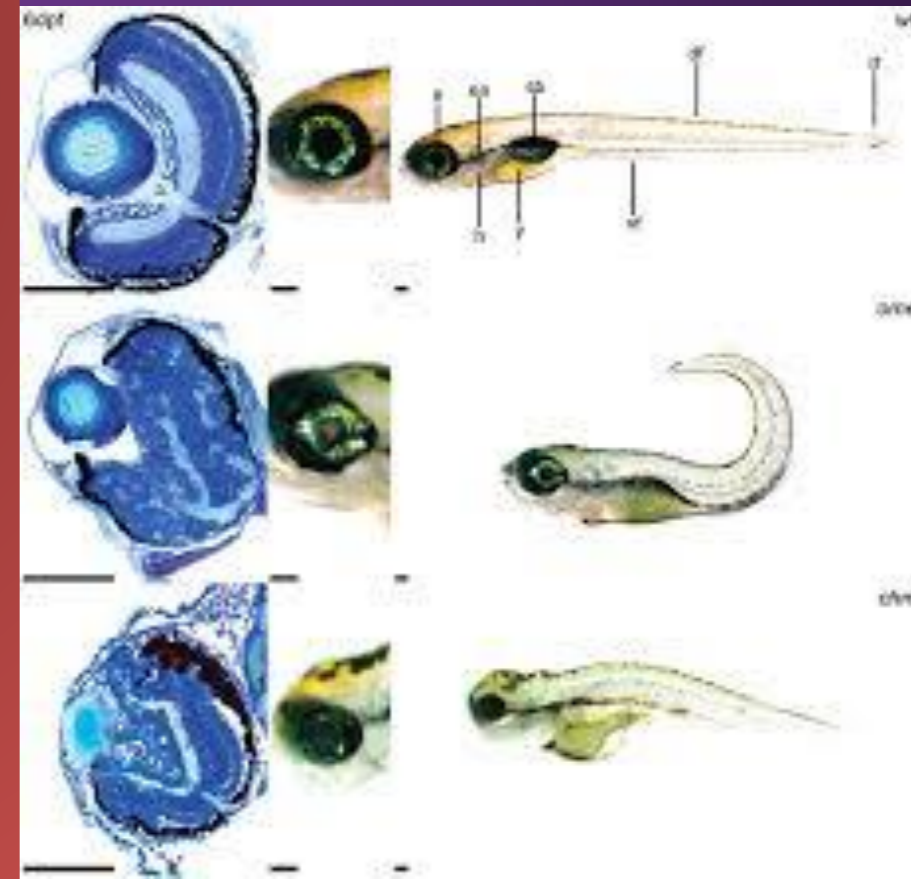
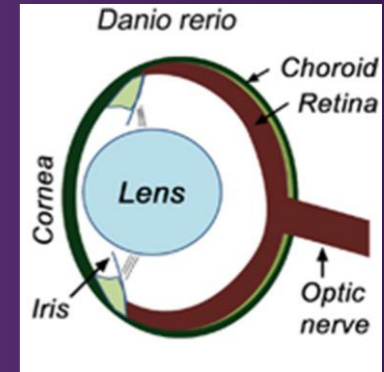
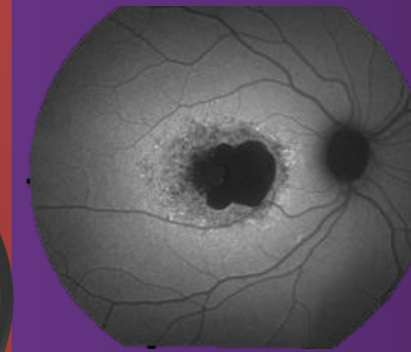


Species/Abbrev	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1. ENSDARP00000018379	M	G	F	A	R	Q	I	K	L	L	L	W	K	N	V	
2. ENSRNOP00000017878	M	G	F	L	R	Q	I	Q	L	L	L	W	K	N	V	
3. ENSMFAP00000018204	M	G	F	V	R	Q	I	Q	L	L	L	W	K	N	V	
4. ENSPPAP00000024958	M	G	F	V	R	Q	I	Q	L	L	L	W	K	N	V	
5. ENSPTRP00000062840	M	G	F	V	R	Q	I	Q	L	L	L	W	K	N	V	
6. ENSDARP000000113256	M	S	T	G	R	Q	I	R	L	L	L	W	K	N	V	
7. ENSDARP000000123162	M	G	T	N	S	Q	V	R	L	L	L	W	K	N	V	
8. ENSGALP00000009224	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	
9. ENSBTAP000000023982	M	G	F	A	R	Q	I	K	L	L	L	W	K	N	V	
10. ENSFCAP00000001439	M	G	F	V	R	Q	I	Q	L	L	L	W	K	N	V	
11. ENSECAP00000001236	M	G	F	A	R	Q	I	Q	L	L	L	W	K	N	V	
12. ENSCAFP00000000497	M	G	F	A	R	Q	I	Q	L	L	L	W	K	N	V	
13. ENSOCUP00000000845	M	G	F	A	R	Q	I	Q	L	L	L	W	K	N	V	
14. ENSSSCP00000000734	M	G	F	A	R	Q	I	R	L	L	L	W	K	N	V	
15. ENSMUSP00000001399	M	G	F	L	R	Q	I	Q	L	L	L	W	K	N	V	
16. ENSP0000000359245	M	G	F	V	R	Q	I	Q	L	L	L	W	K	N	V	

## Develop mutants

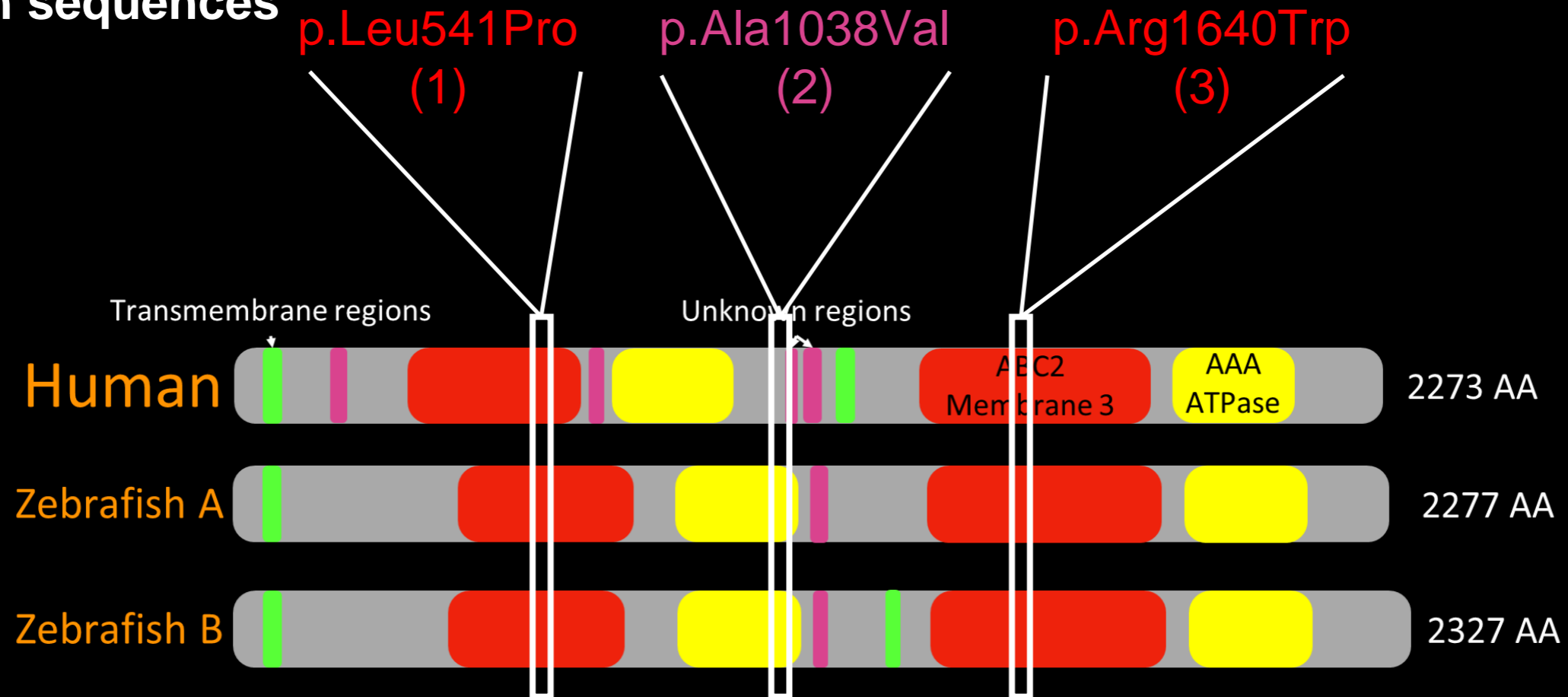


## Screen phenotypes



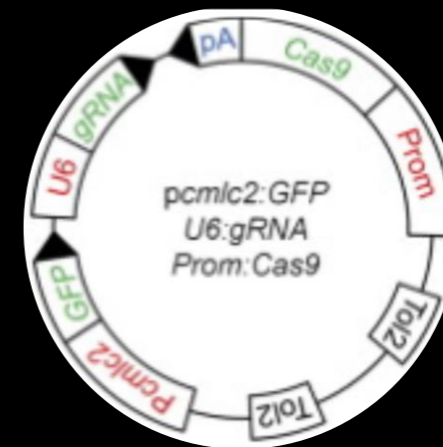
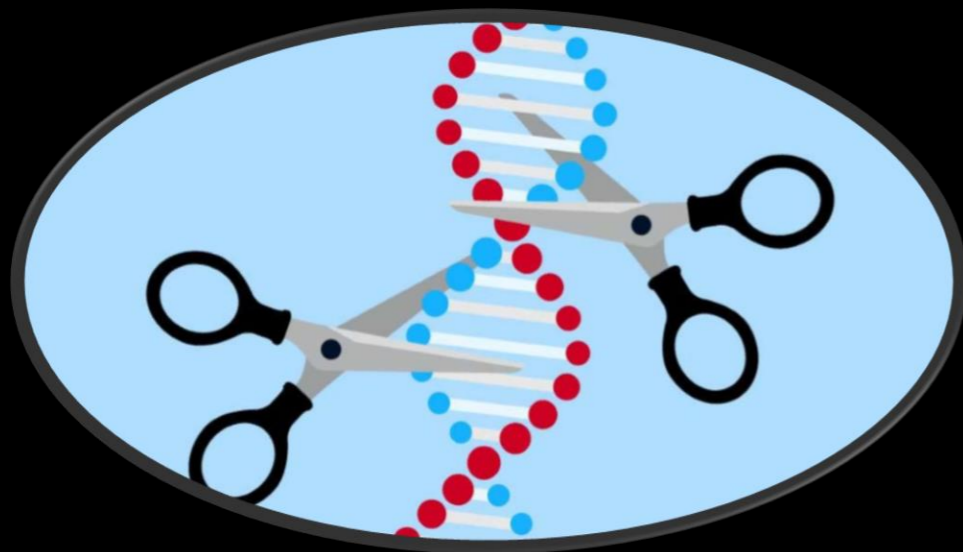
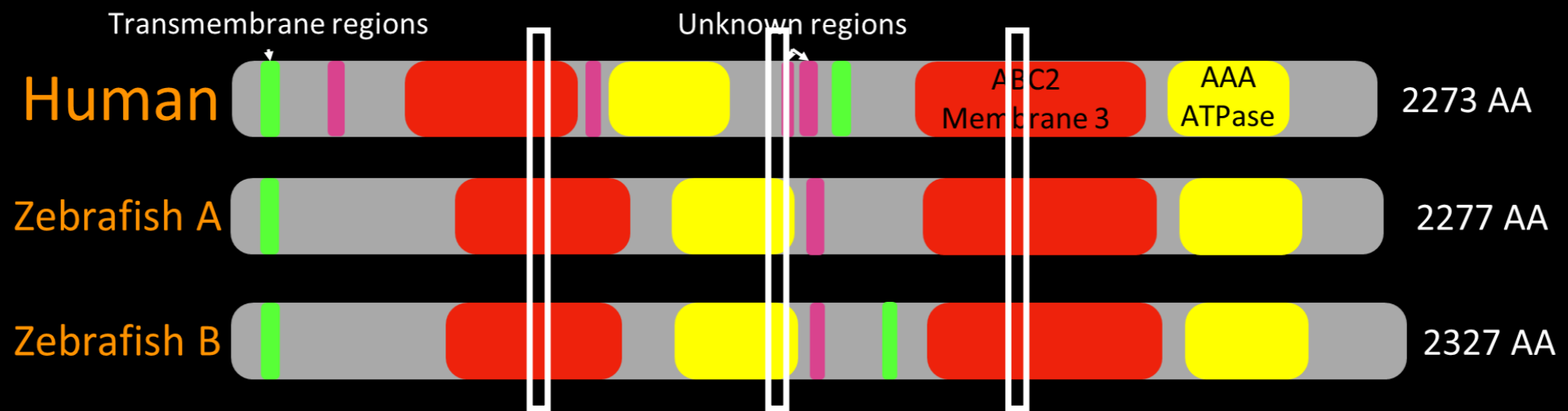
# Aim 1: Determine domains important for retinol metabolism throughout development

Align sequences



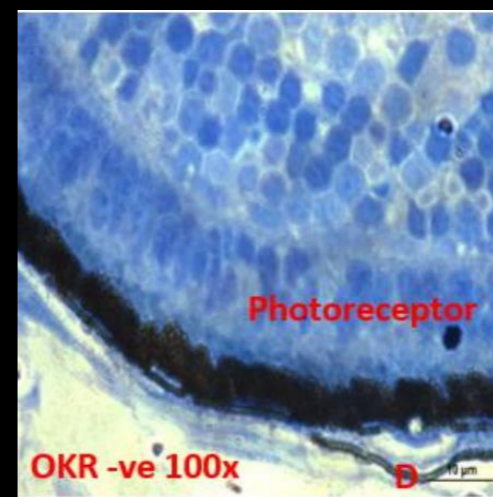
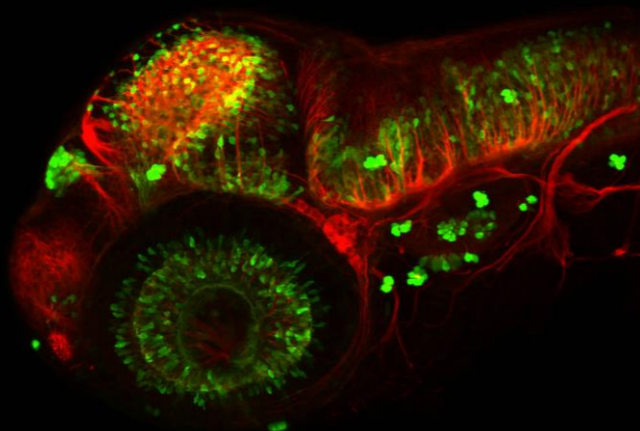
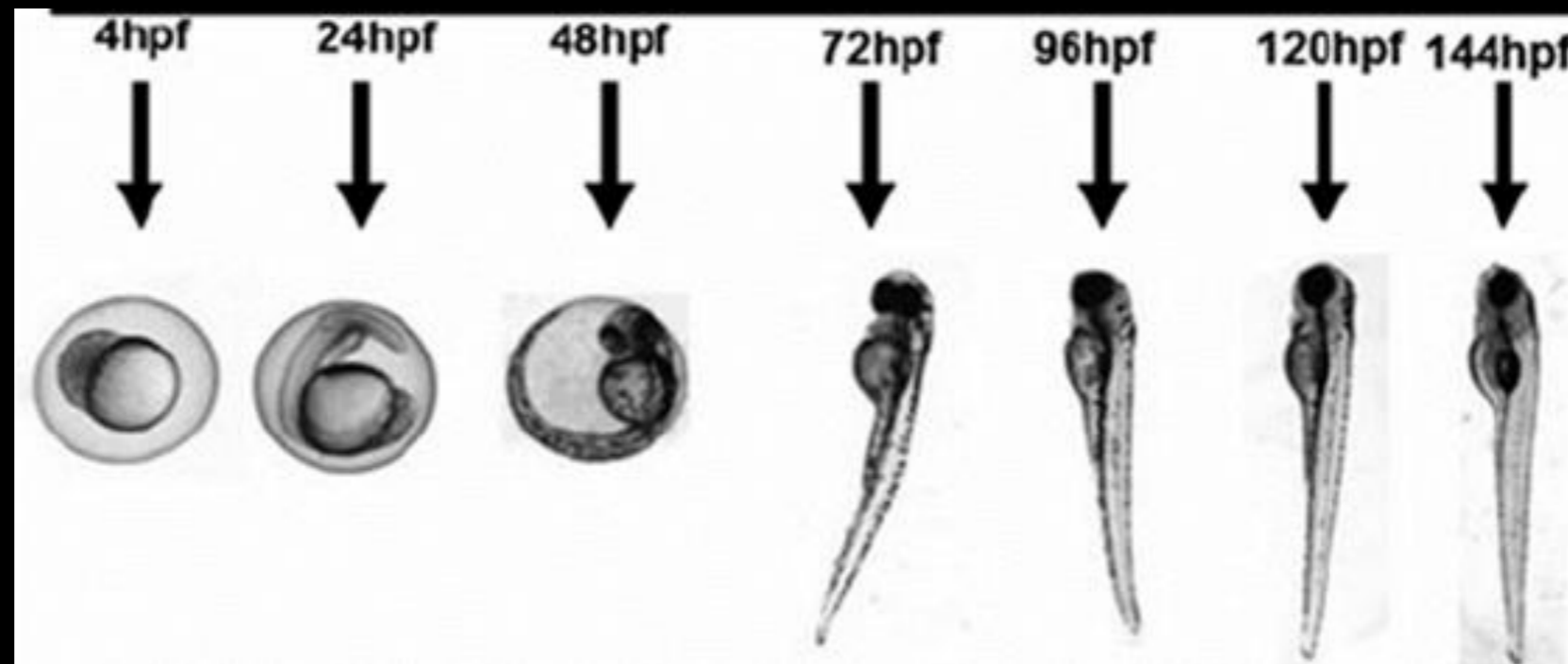
# Aim 1: Determine domains important for retinol metabolism throughout development

## Develop mutants



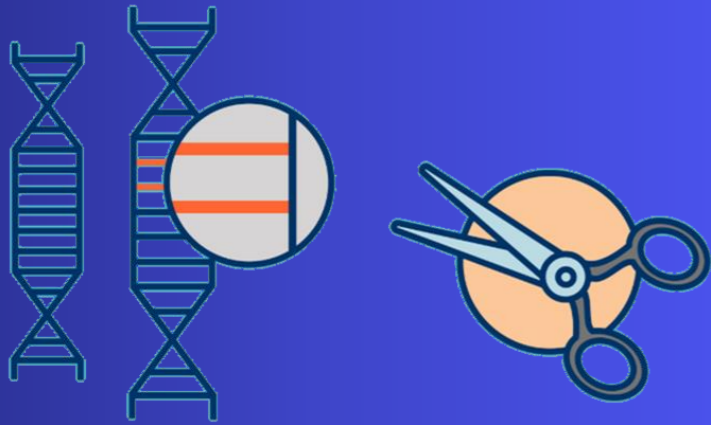
# Aim 1: Determine domains important for retinol metabolism throughout development

## Screen phenotypes



# Aim 2: Identify chemical modulation of retinol metabolism during development

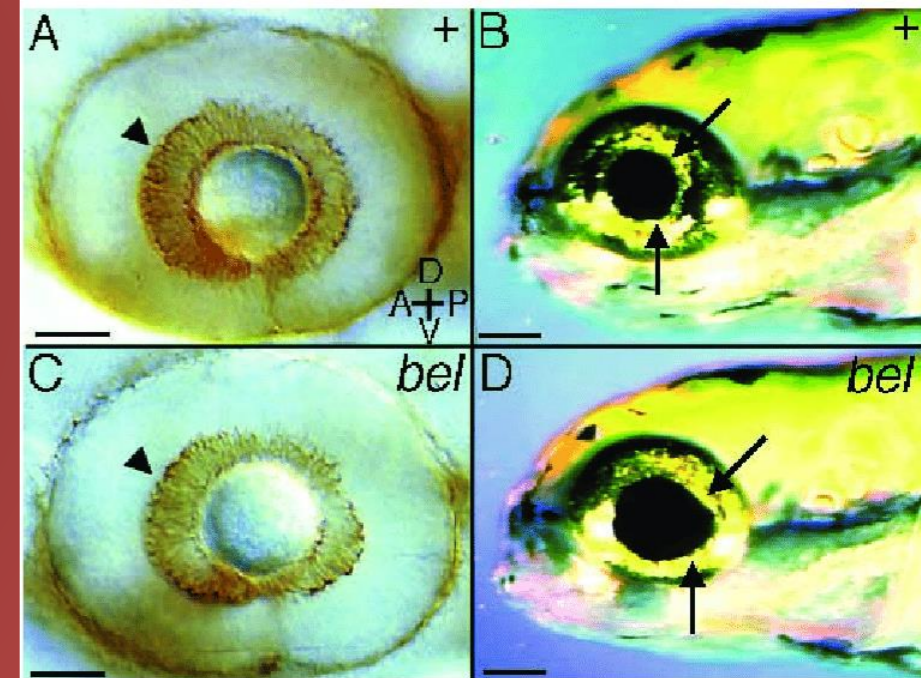
Develop mutant and WT ABCA4 populations



Perform chemical screen



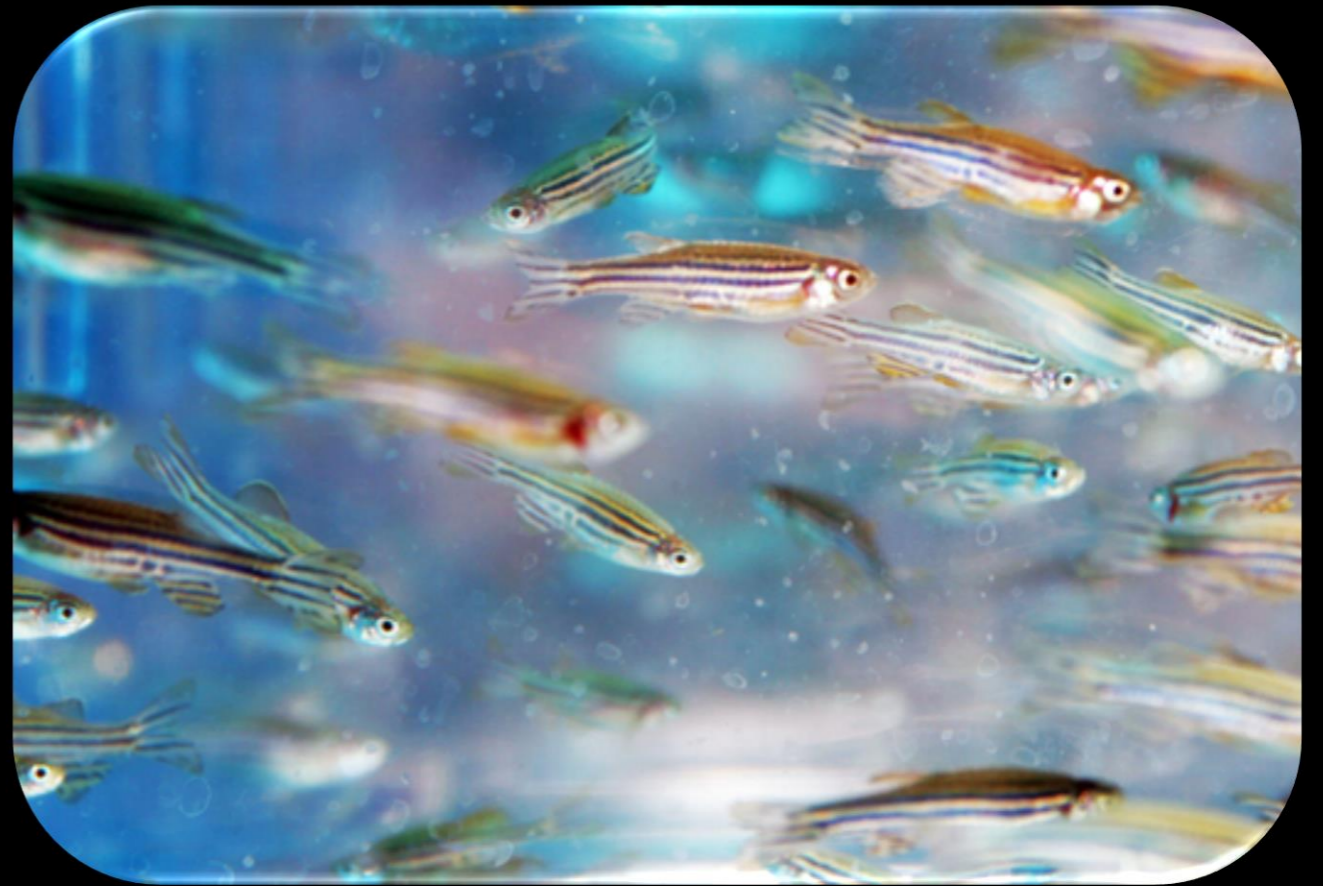
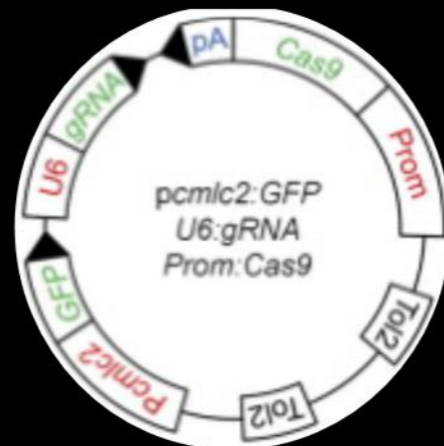
Analyze resulting phenotypes



# Aim 2: Identify chemical modulation of retinol metabolism during development

Develop mutant and WT ABCA4 populations

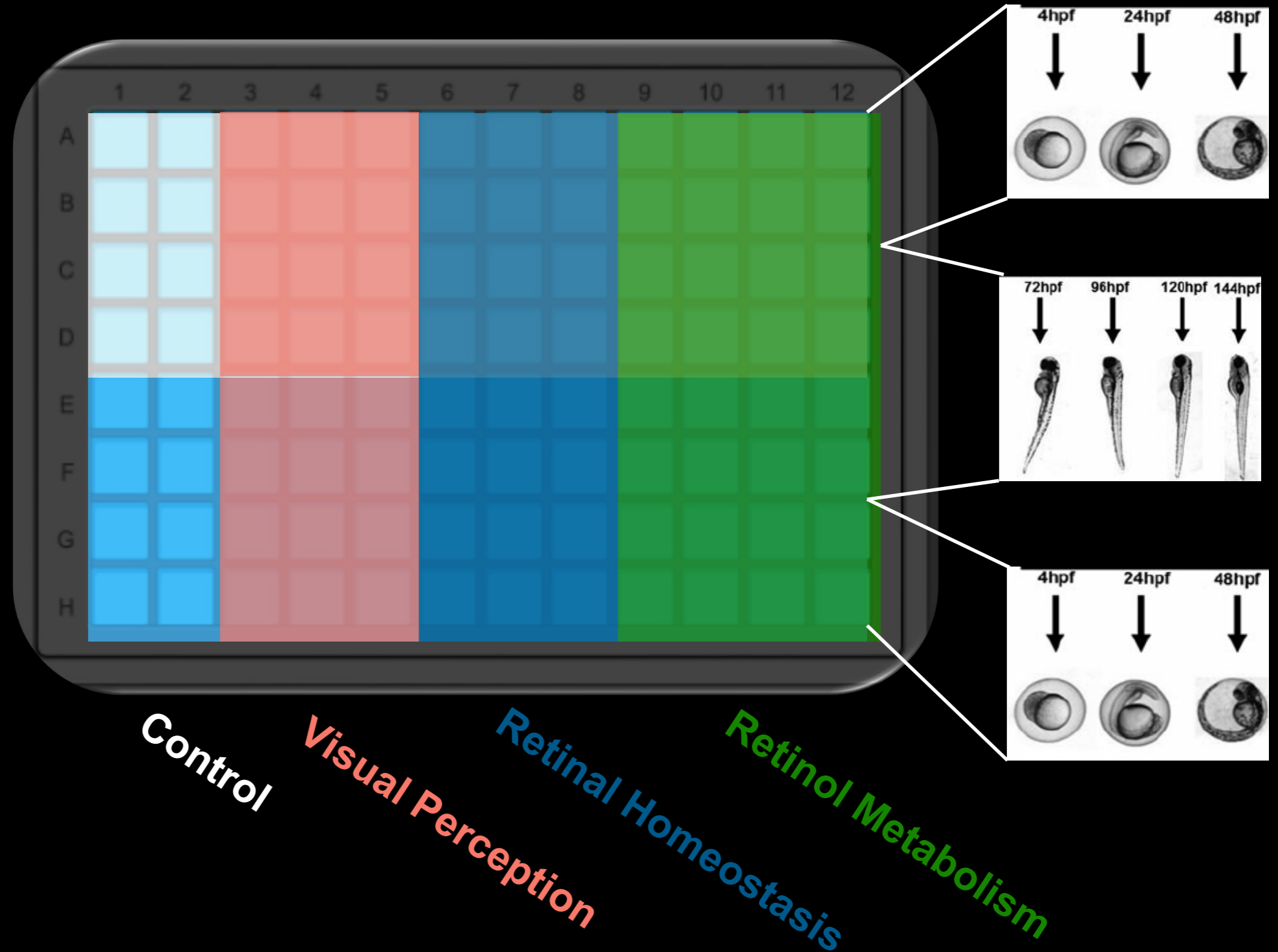
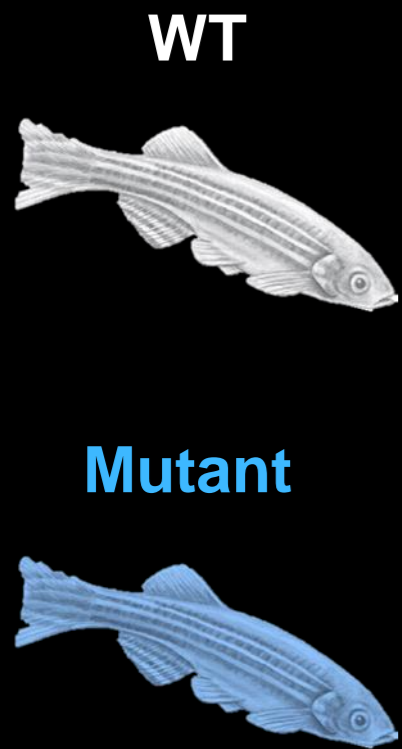
- p.Leu541Pro (1)
- p.Ala1038Val (2)
- p.Arg1640Trp (3)





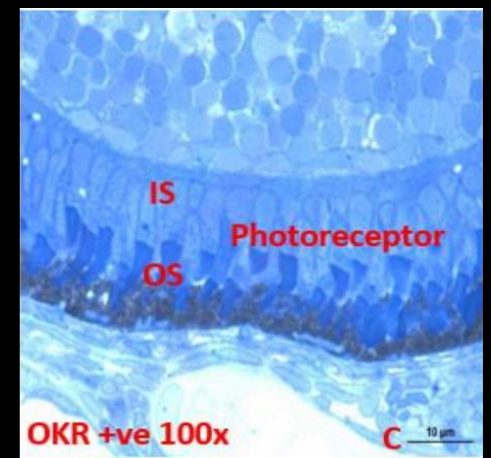
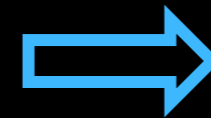
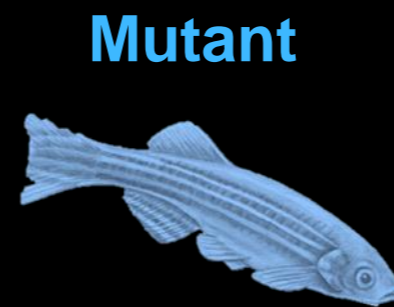
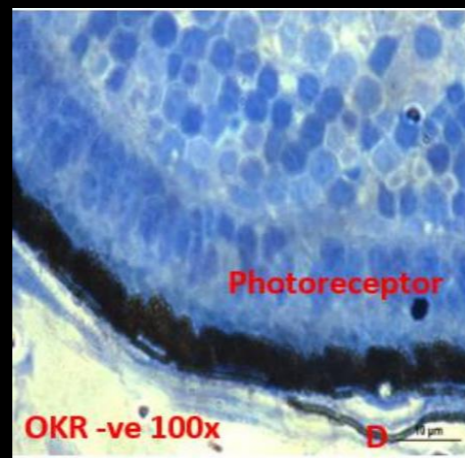
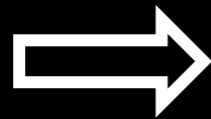
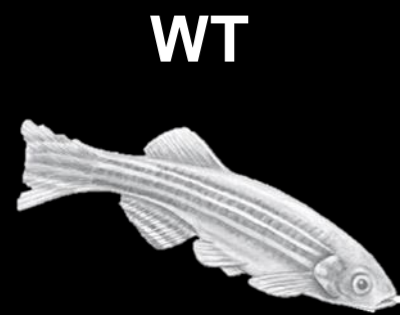
# Aim 2: Identify chemical modulation of retinol metabolism during development

Perform chemical screen



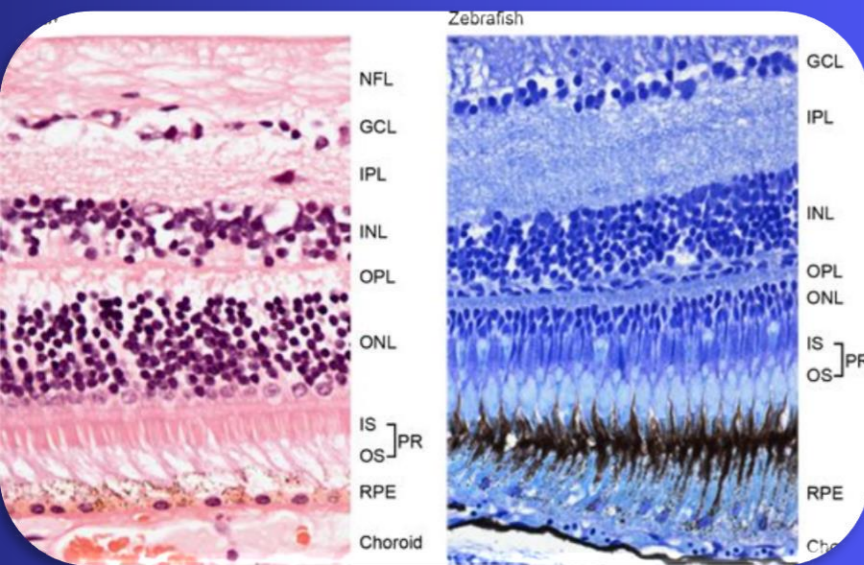
# Aim 2: Identify chemical modulation of retinol metabolism during development

Analyze resulting phenotypes

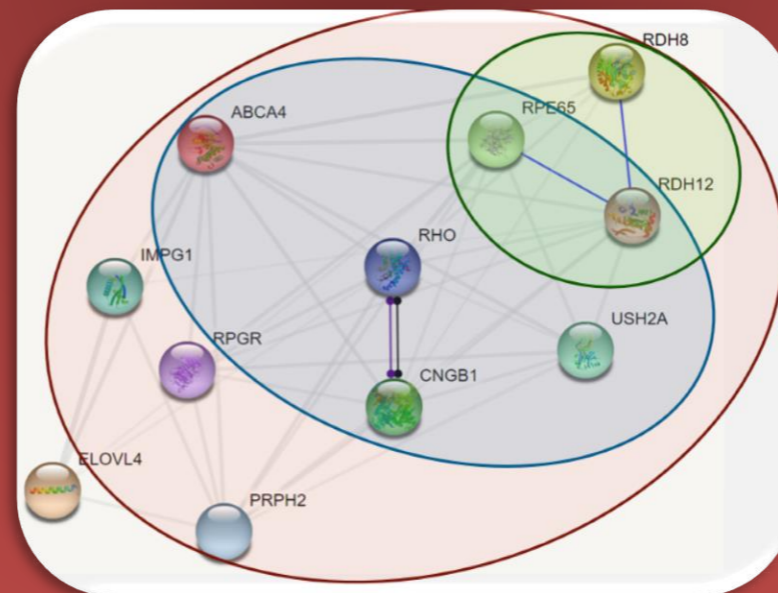
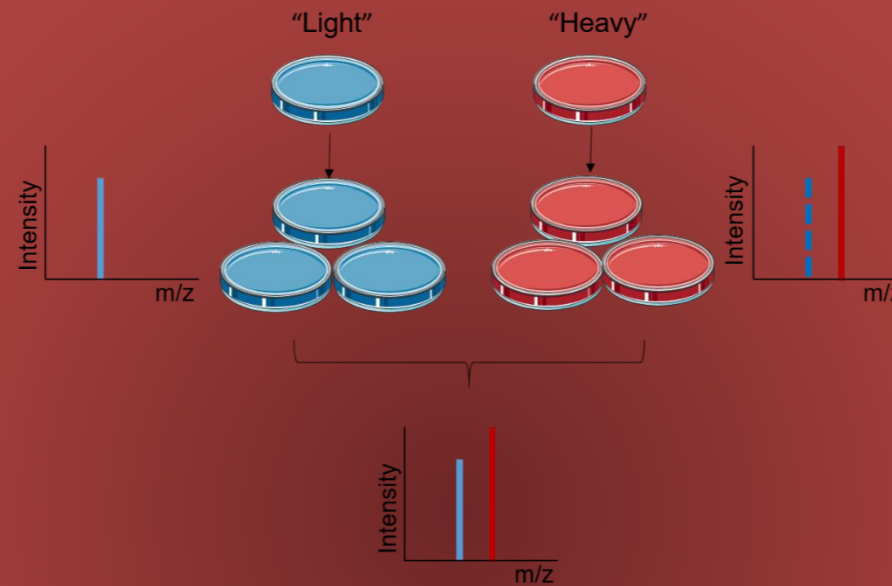


# Aim 3: Determine the expression of retinol metabolism genes through time

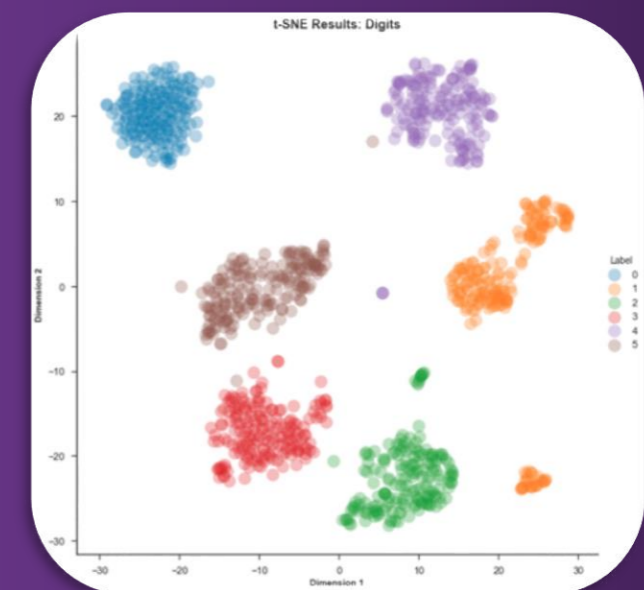
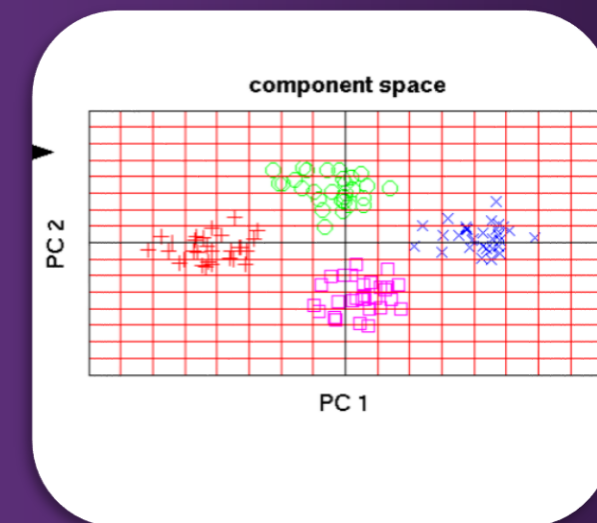
## Isolate cells



## Pulsed SILAC



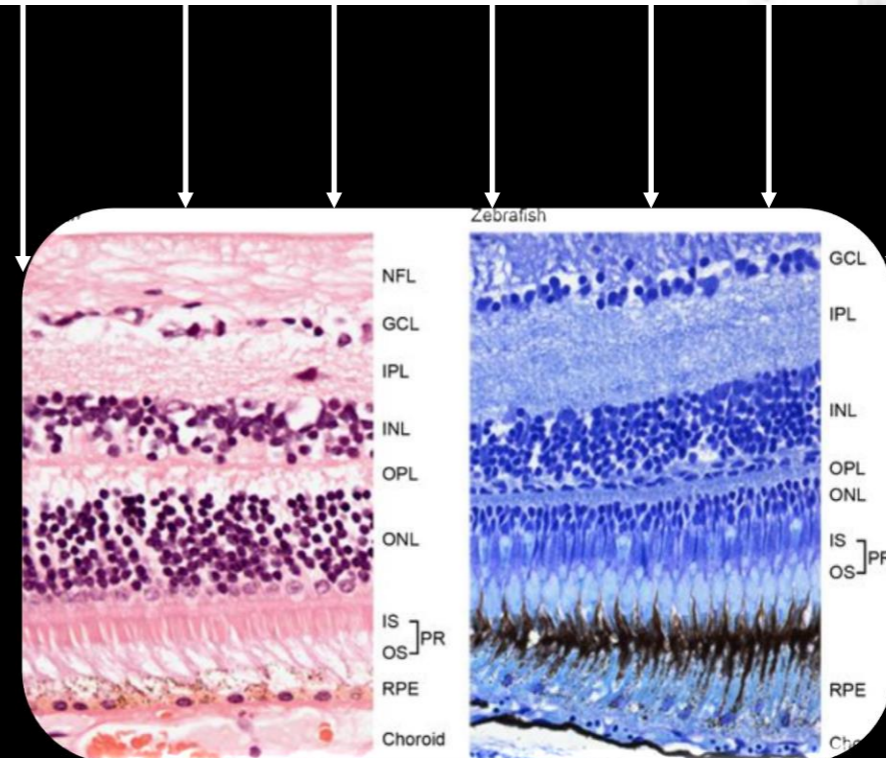
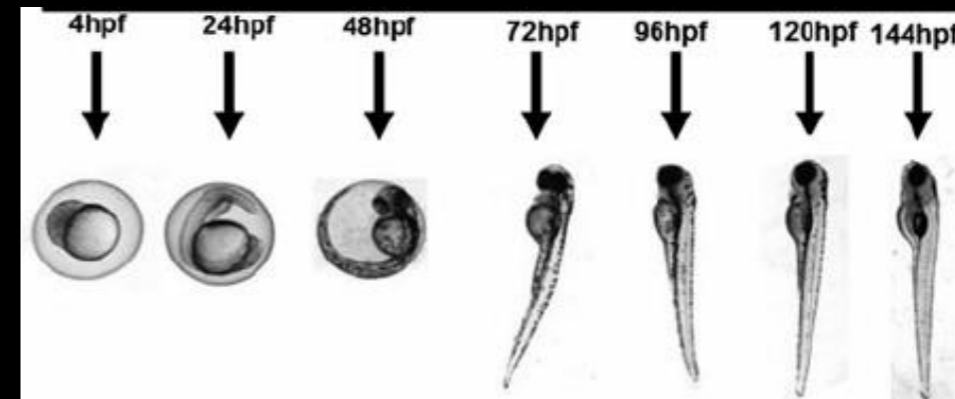
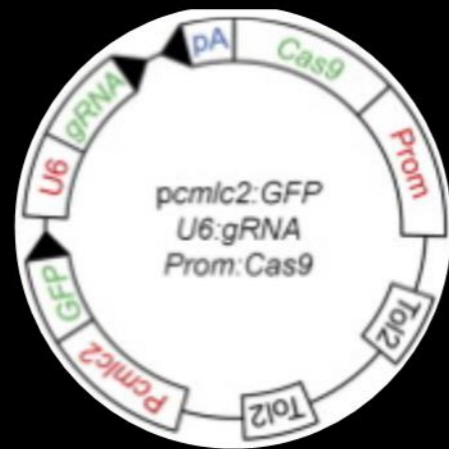
## Dimensional reduction and analysis



# Aim 3: Determine the expression of retinol metabolism genes through time

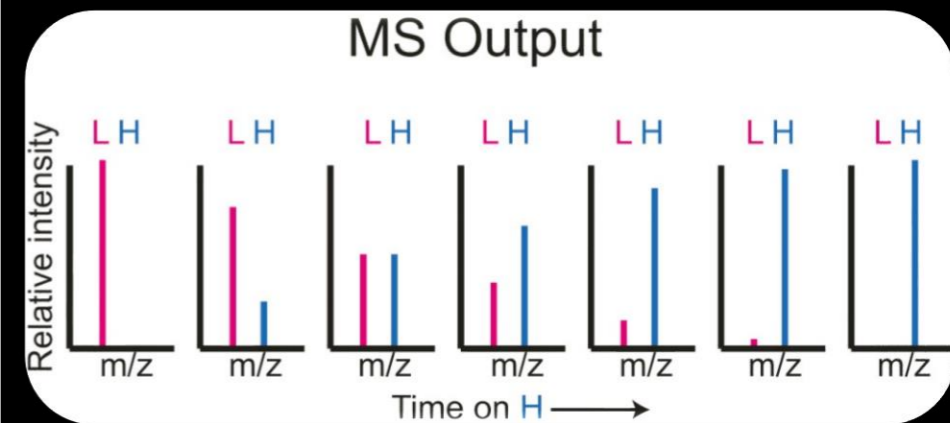
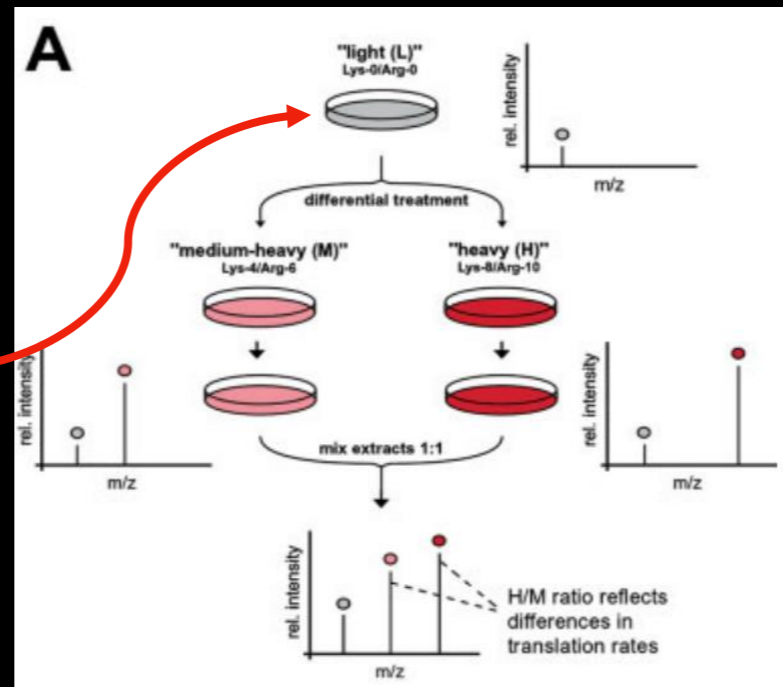
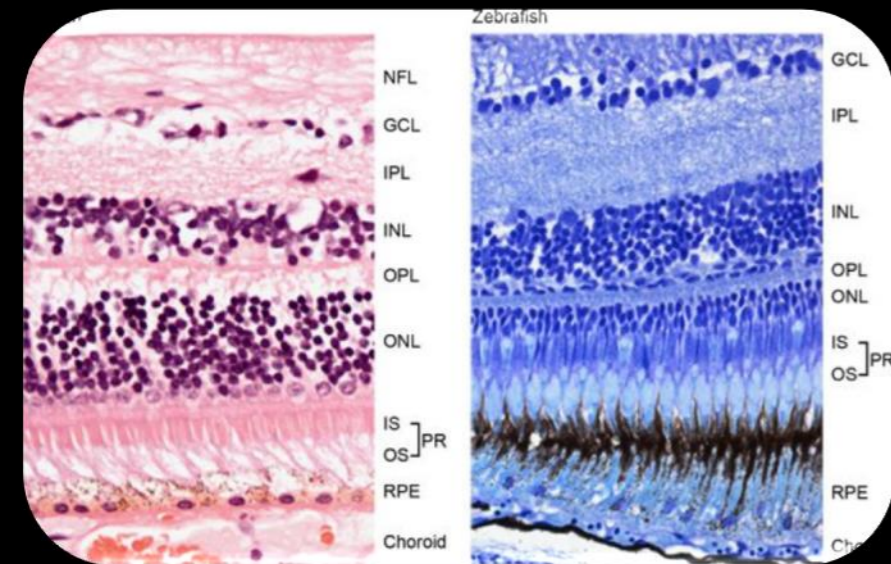
## Isolate cells

- p.Leu541Pro (1)
- p.Ala1038Val (2)
- p.Arg1640Trp (3)



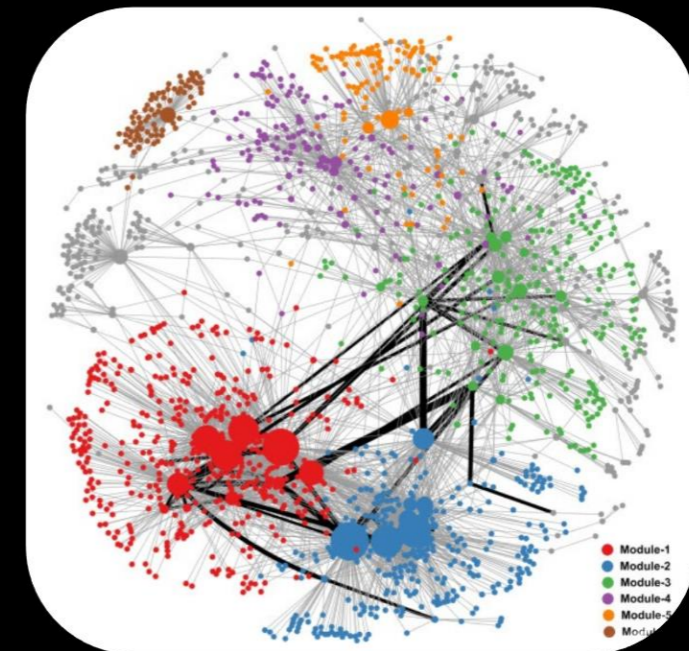
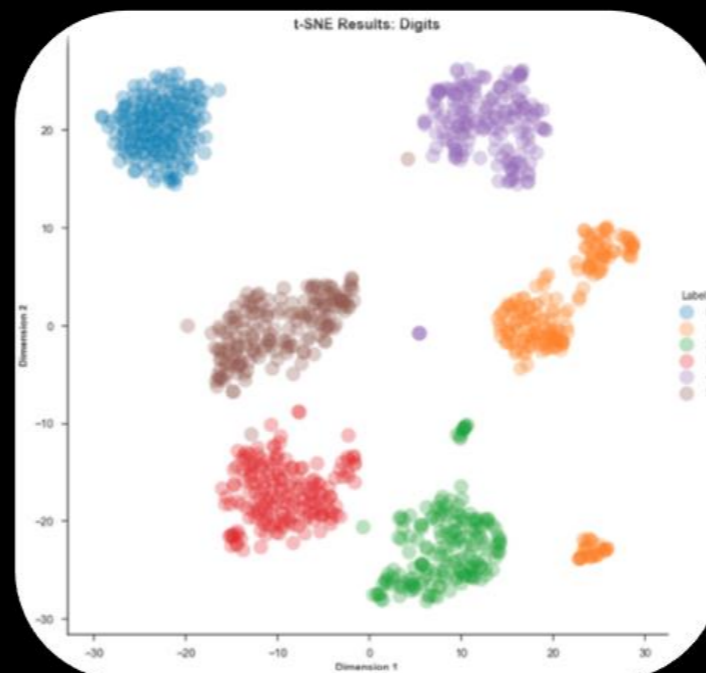
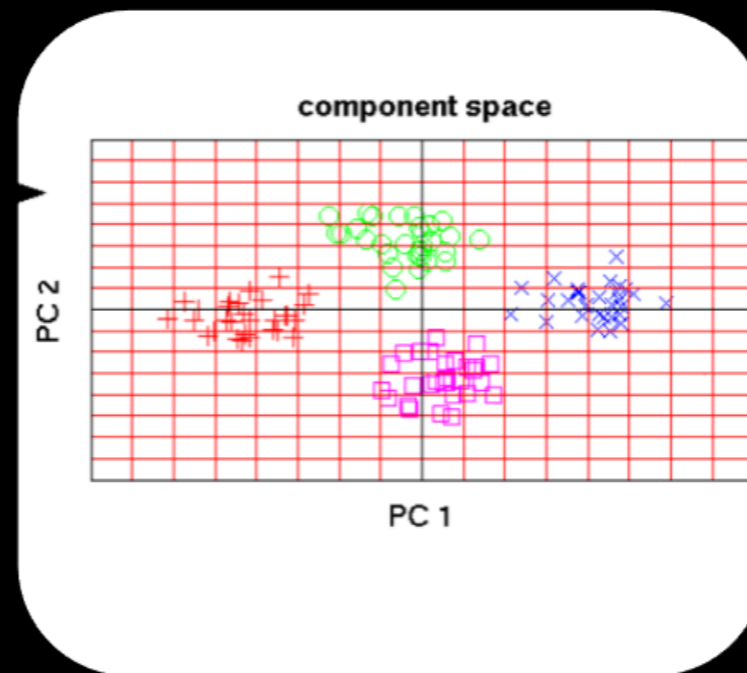
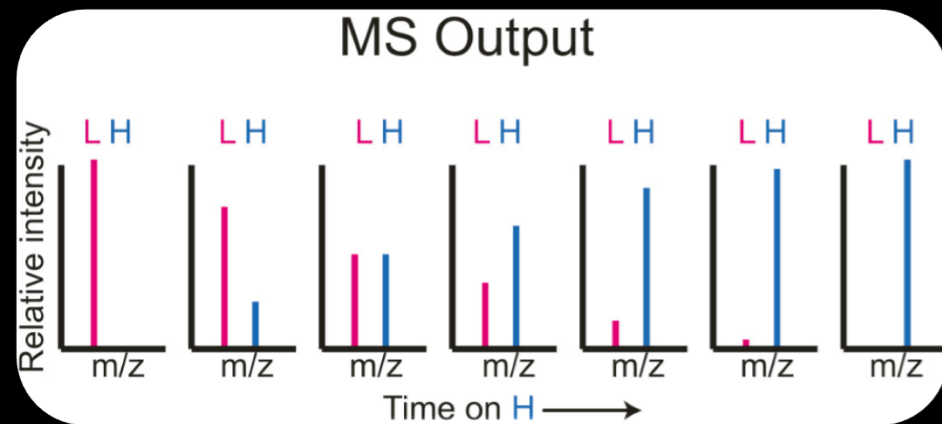
# Aim 3: Determine the expression of retinol metabolism genes through time

## Pulsed SILAC



# Aim 3: Determine the expression of retinol metabolism genes through time

## Dimensional reduction and analysis

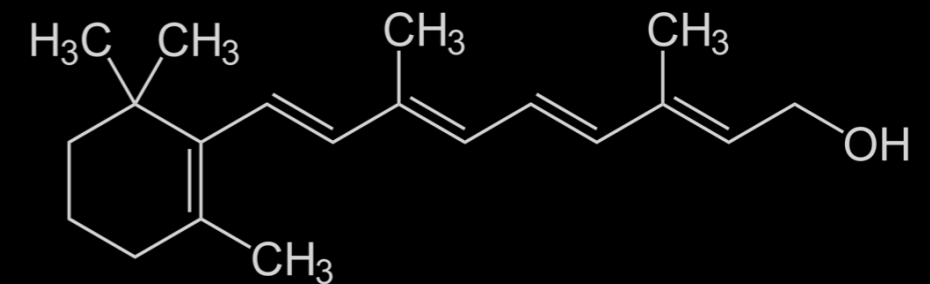


# Conclusion

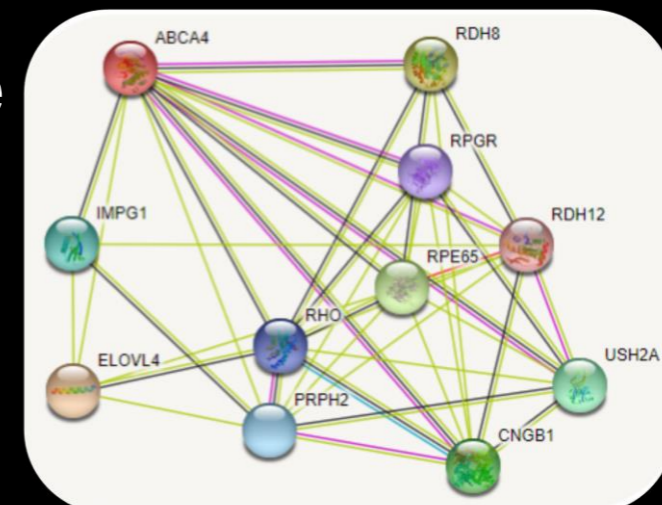
Alignment of sequences and determination of mutant Zebrafish phenotypes may assist with understanding modes of retinol metabolism differing between species.

Protein Sequences	
Species/Abbrv	
1. ENSOARP00000018379	M G F A R Q I K L L L W K N V
2. ENSRNOF00000017878	M G F L R Q I Q L L L W K N V
3. ENSMFAP00000018204	M G F V R Q I Q L L L W K N V
4. ENSPPAP00000024958	M G F V R Q I Q L L L W K N V
5. ENSPTRP000000062840	M G F V R Q I Q L L L W K N V
6. ENSDARP000000113256	M S T G R Q I R L L L W K N V
7. ENSDIAR000000123162	M G T N S Q V R L L L W K N V
8. ENSGALP00000009224	
9. ENSBTAP00000023982	M G F A R Q I K L L L W K N V
10. ENSFCAP0000001439	M G F V R Q I Q L L L W K N V
11. ENSECAP0000001236	M G F A R Q I Q L L L W K N V
12. ENSCAFP0000000497	M G F A R Q I Q L L L W K N V
13. ENSOCUP00000000845	M G F A R Q I Q L L L W K N V
14. ENSSSCP00000007345	M G F A R Q I R L L L W K N V
15. ENSMUSP0000001399	M G F L R Q I Q L L L W K N V
16. ENSP000000359245	M G F V R Q I Q L L L W K N V

Performing chemical screens on wildtype and mutant Zebrafish can elucidate chemical modulation of retinol metabolism.



Understanding the expression of cells in the retina can further the understanding of ABCA4 interactions in retinol metabolism.



# Future Directions





Questions?

# Image References

<https://130529218-701413549797869494.preview.editmysite.com/uploads/1/3/0/5/130529218/599754248.jpg>

<https://www.centervue.com/wp-content/uploads/2017/07/image1.jpg>

[https://completeeyecare.com/wp-content/uploads/2013/09/SKMBT\\_C36013111815130-300x217.jpg](https://completeeyecare.com/wp-content/uploads/2013/09/SKMBT_C36013111815130-300x217.jpg)

<https://cdn.arstechnica.net/wp-content/uploads/2017/03/retina-age-related-macular-degeneration-600x538.png>

<https://media.npr.org/assets/img/2016/03/02/amd-b2ba911d887ba542c6ae64d84ab008d7f0992d8b-s800-c85.jpg>

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<https://c7.uihere.com/files/980/612/970/human-eye-diagram-eye-pattern-eye.jpg>

<https://www.nei.nih.gov/sites/default/files/styles/large/public/2019-05/Stargardtiseasefundusautofluorescence-Zein.jpg?itok=U7U-G7VT>

<https://paintingvalley.com/drawings/zebrafish-drawing-13.jpg>

[https://www.researchgate.net/profile/Jamuna\\_Chhetri/publication/260120244/figure/fig1/AS:279070190391347@1443546926399/Comparison-of-the-human-and-zebrafish-eye-a-Comparison-of-the-human-and-zebrafish-eye.png](https://www.researchgate.net/profile/Jamuna_Chhetri/publication/260120244/figure/fig1/AS:279070190391347@1443546926399/Comparison-of-the-human-and-zebrafish-eye-a-Comparison-of-the-human-and-zebrafish-eye.png)

[https://www.researchgate.net/profile/Iain\\_Shepherd/publication/26302833/figure/fig1/AS:277082430033942@1443073007999/Life-cycle-of-the-zebrafish-Zebrafish-develop-rapidly-from-a-one-cell-zygote-that-sits.png](https://www.researchgate.net/profile/Iain_Shepherd/publication/26302833/figure/fig1/AS:277082430033942@1443073007999/Life-cycle-of-the-zebrafish-Zebrafish-develop-rapidly-from-a-one-cell-zygote-that-sits.png)

<https://www.pnas.org/content/pnas/102/2/407/F5.large.jpg>

<https://paintingvalley.com/sketches/simple-human-body-sketch-26.jpg>

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[https://www.oist.jp/sites/default/files/photos/zebrafish\\_0.jpg](https://www.oist.jp/sites/default/files/photos/zebrafish_0.jpg)

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[https://www.researchgate.net/profile/Bernhard\\_Mueller/publication/14201832/figure/fig3/AS:669217166733315@1536565214149/belladonna-mutant-eye-phenotype-Wild-type-A-and-belladonna-C-eyes-at-48-hours.png](https://www.researchgate.net/profile/Bernhard_Mueller/publication/14201832/figure/fig3/AS:669217166733315@1536565214149/belladonna-mutant-eye-phenotype-Wild-type-A-and-belladonna-C-eyes-at-48-hours.png)