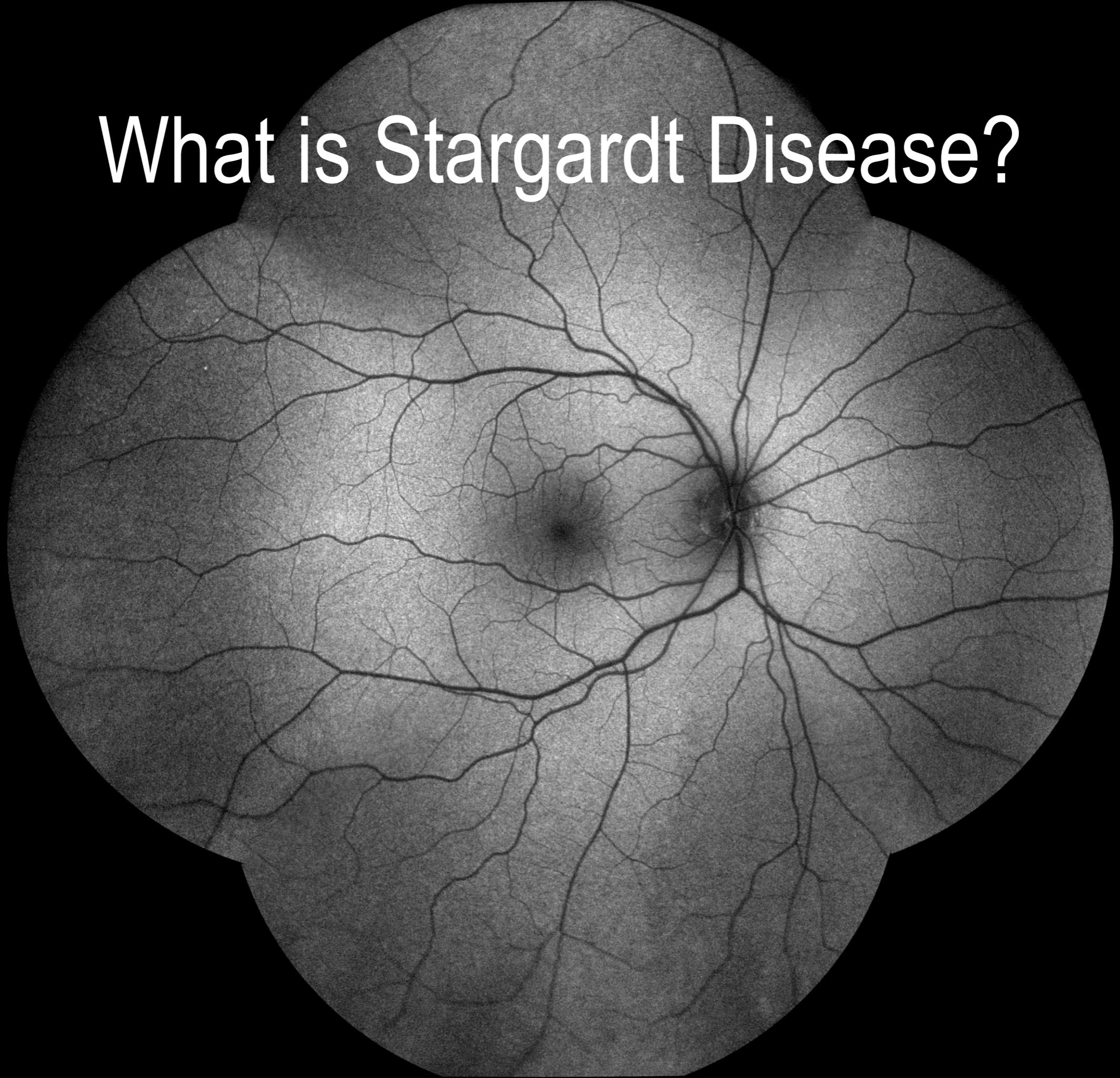


# Inherited Progressive Vision Loss

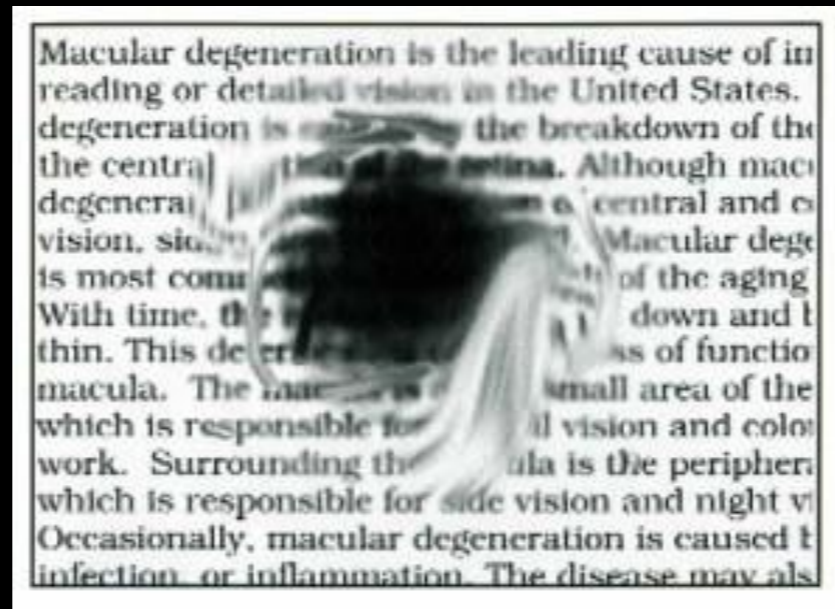
## BCA4 Responsible Stargardt Disease



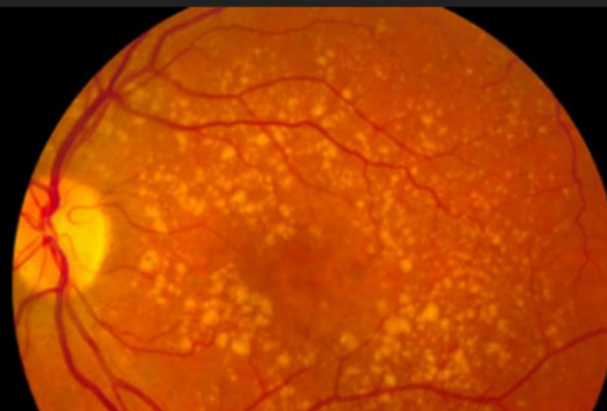
# What is Stargardt Disease?



# What are the Symptoms of Stargardt Disease?



**Loss of central vision**

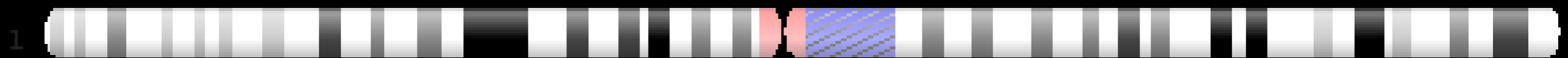


**Yellow-white spots on retina**



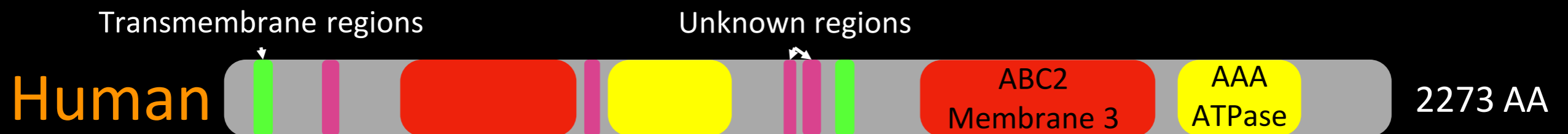
**Colorblindness and eventual complete vision loss**

# ABCA4 is responsible for Stargardt disease

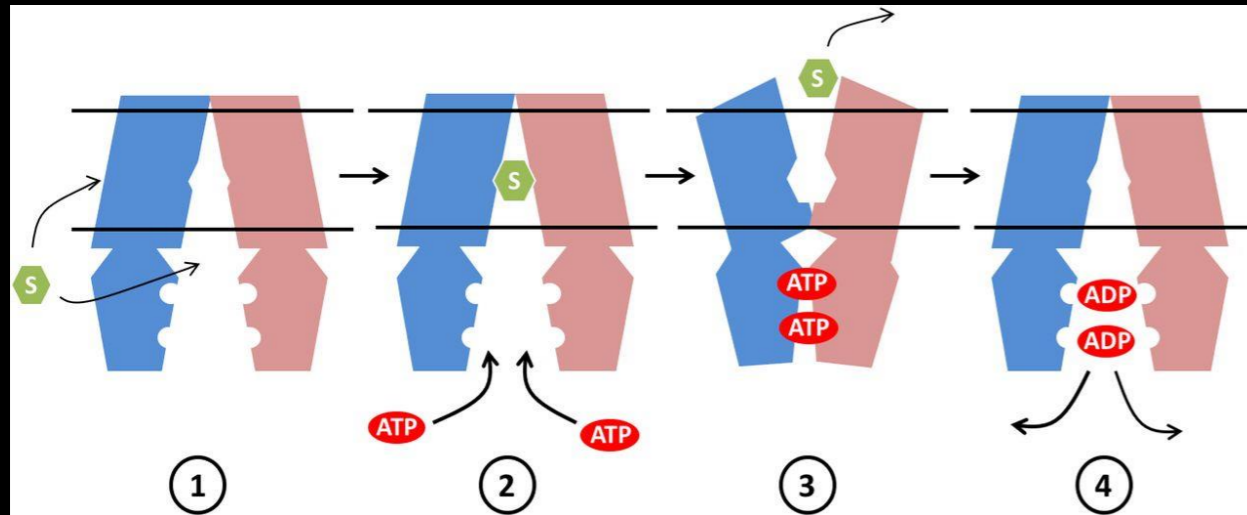


ABCA4

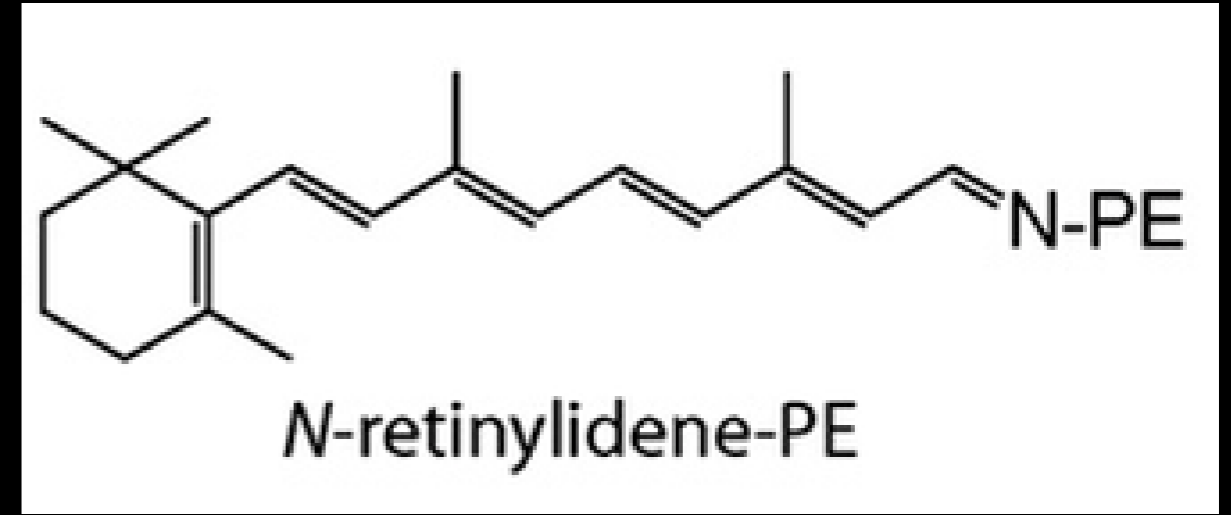
1<sup>st</sup> chromosome, position 22.1



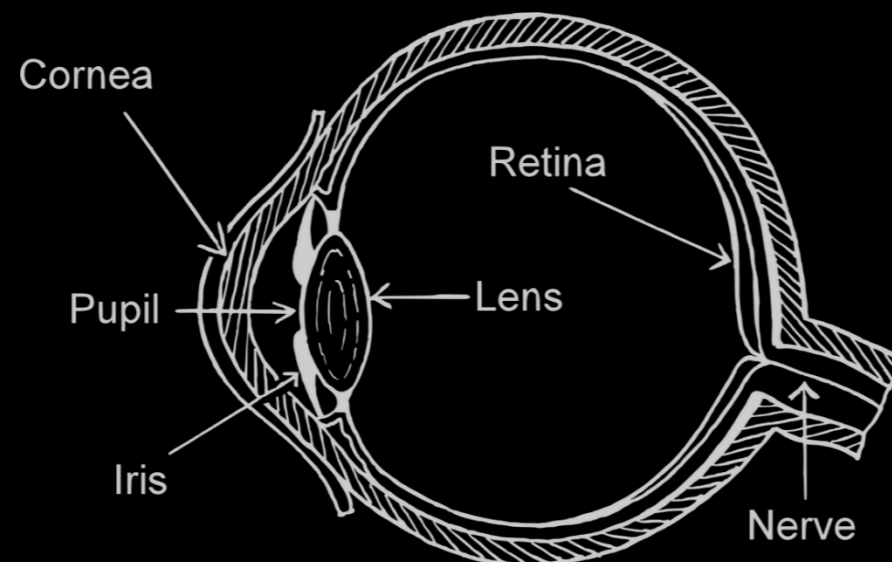
# How does the ABCA4 protein operate?



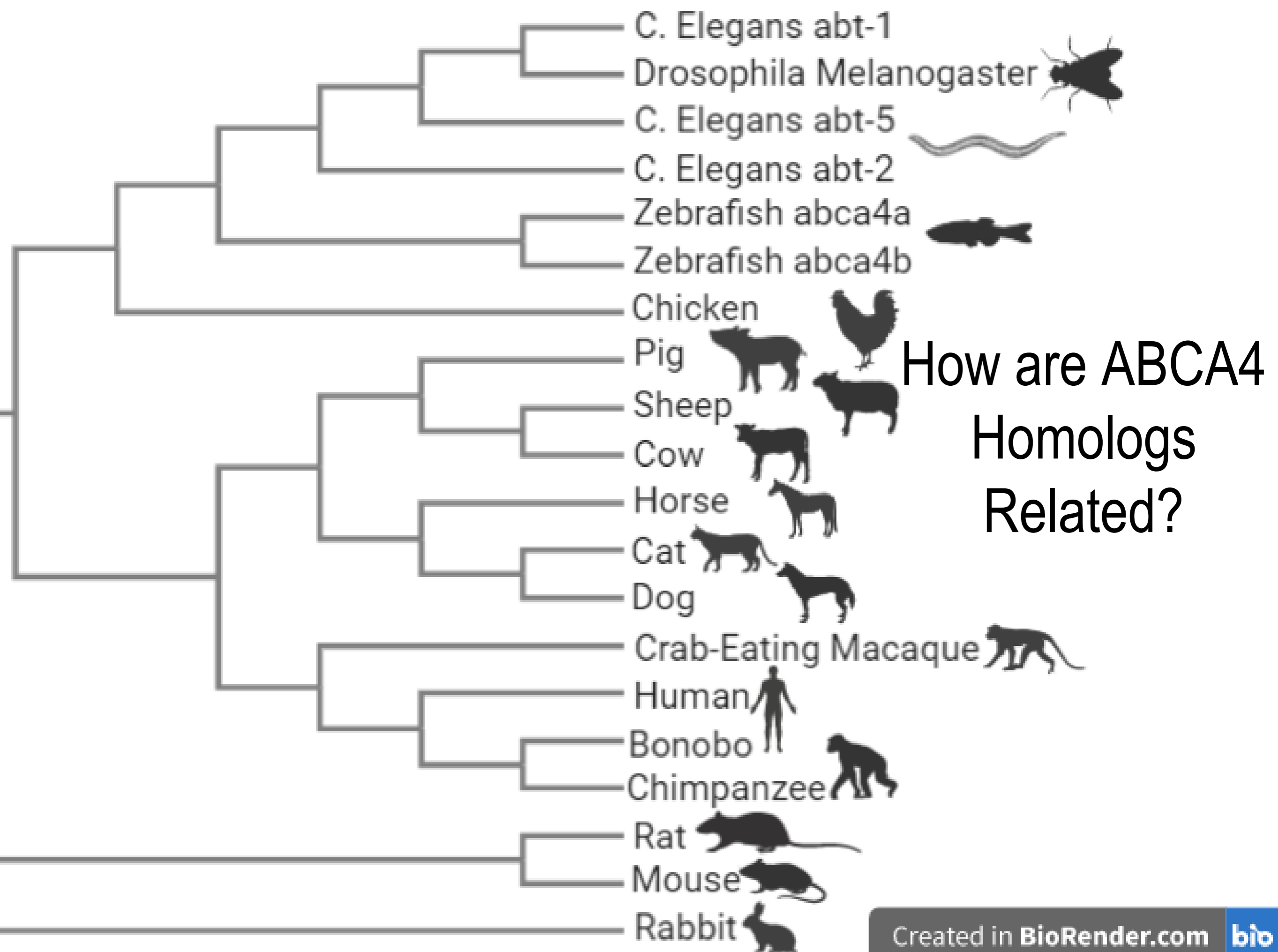
**ATP-Binding Cassette  
Molecular function**



**Lipid Transport on membrane  
Cellular component**



**Visual Perception  
Biological process**



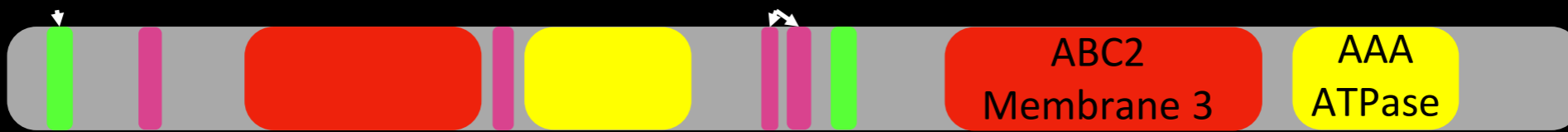
# ABCA4 is conserved between species with...

## Camera-type eyes:

Transmembrane regions

Unknown regions

Human



2273 AA

Bonobo



2159 AA 99%

Mouse



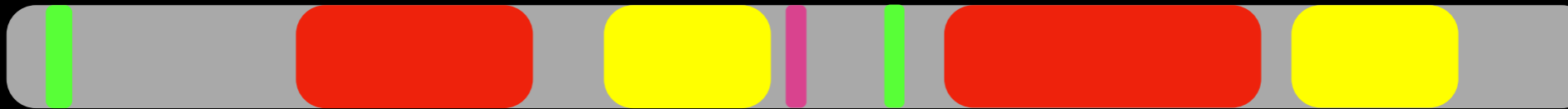
2310 AA 87%

Zebrafish A



2277 AA 63%

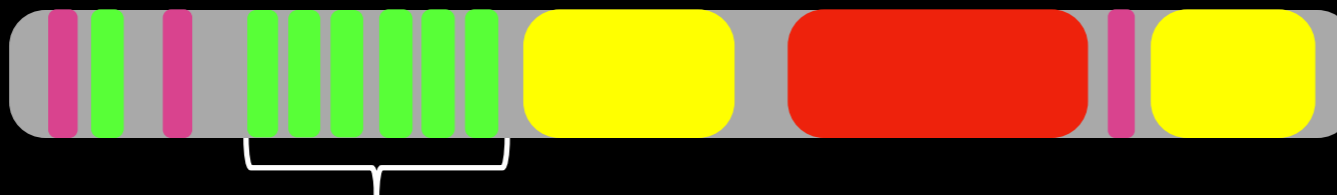
Zebrafish B



2327 AA 63%

## Other eye types:

C. Elegans



1564 AA 27%

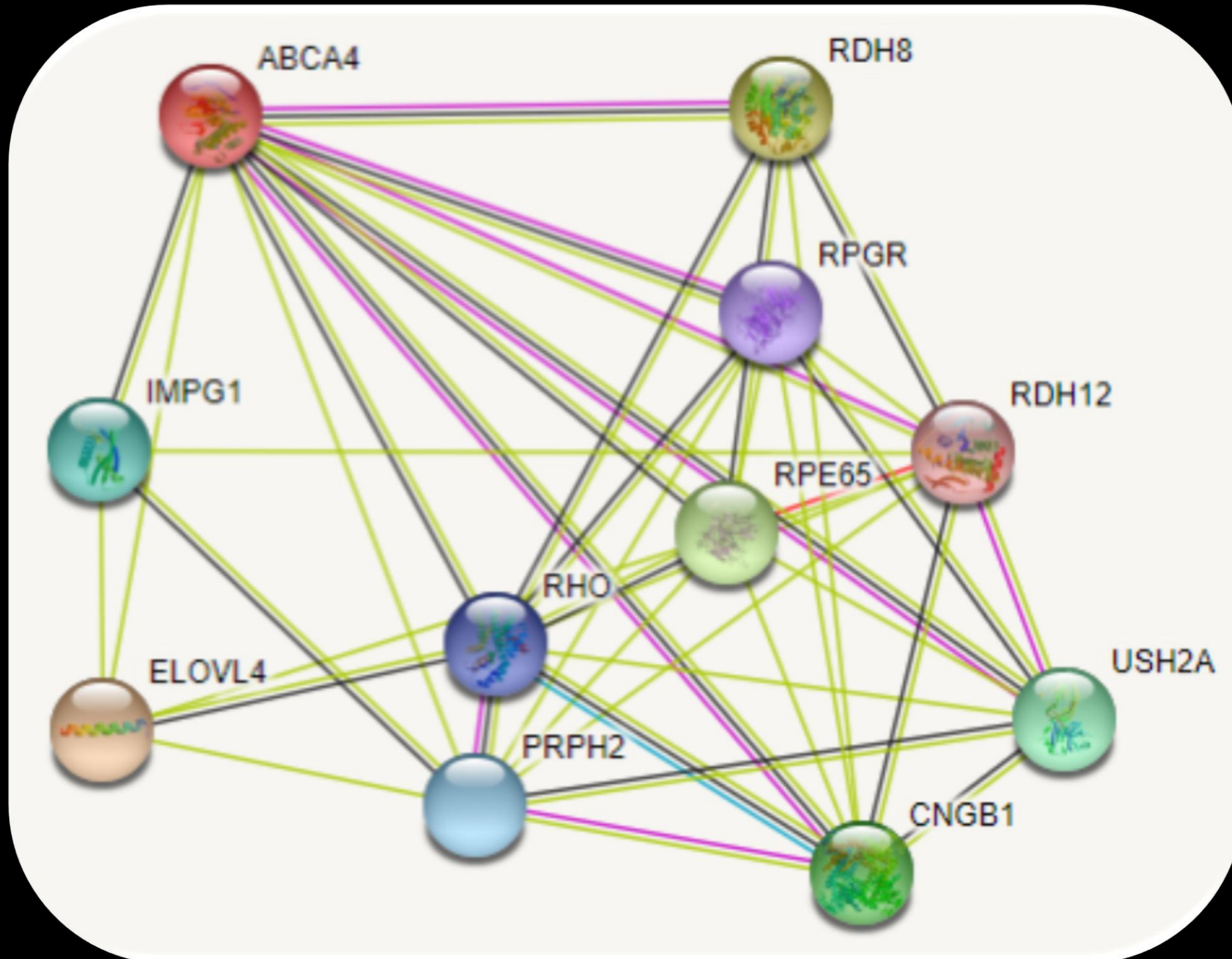
Similar to ABC2 Membrane 3

D. Melanogaster



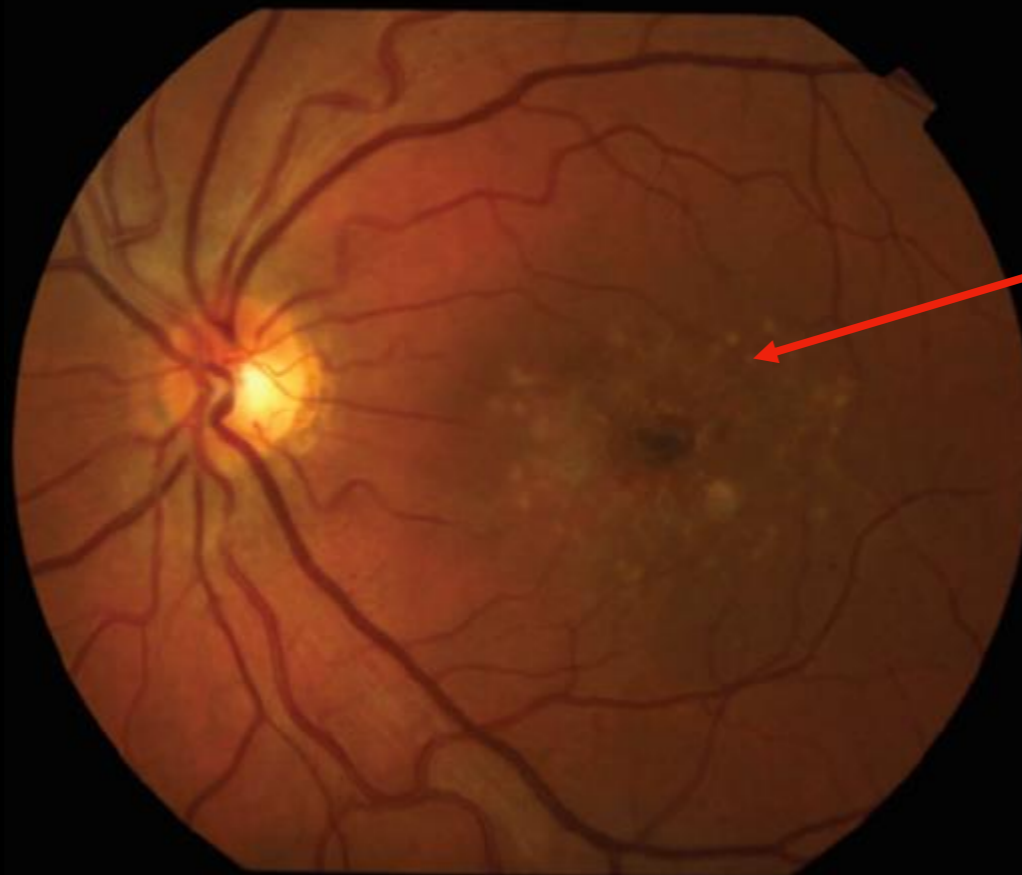
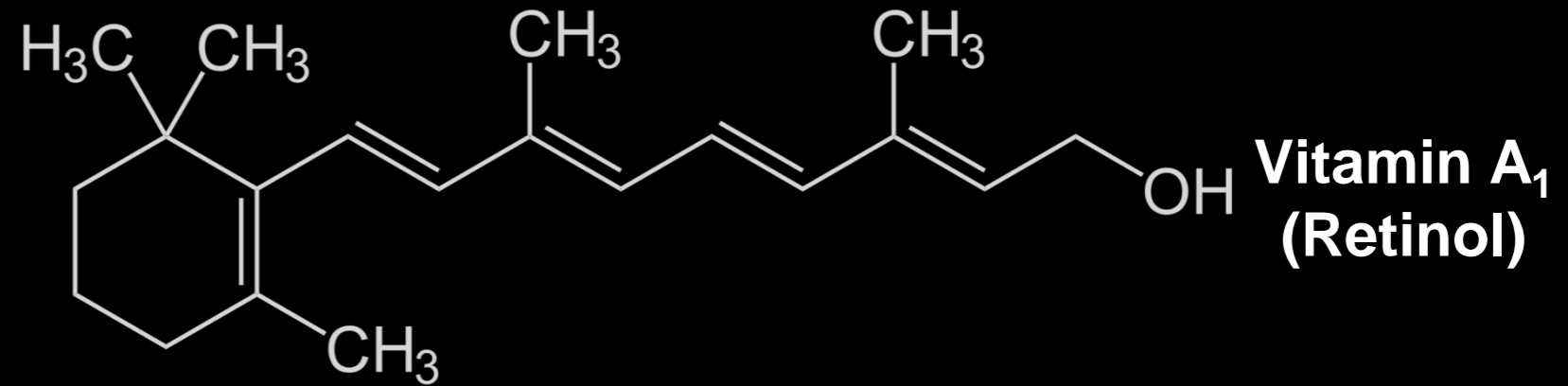
388 AA 29%

# What protein interactions does ABCA4 perform?



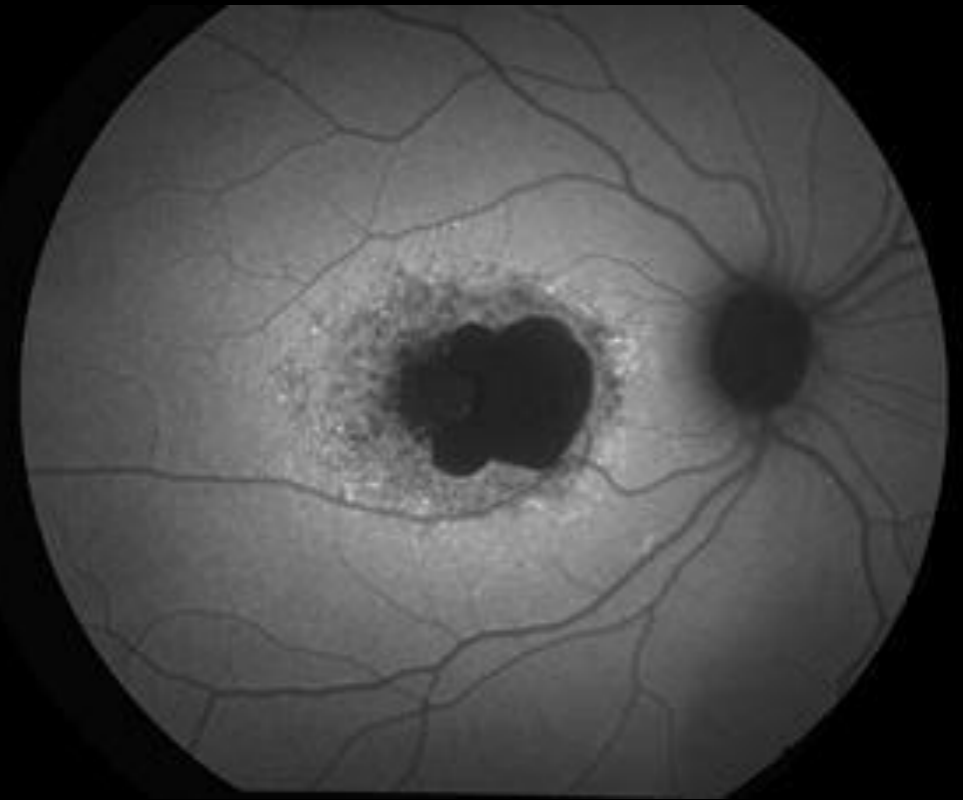
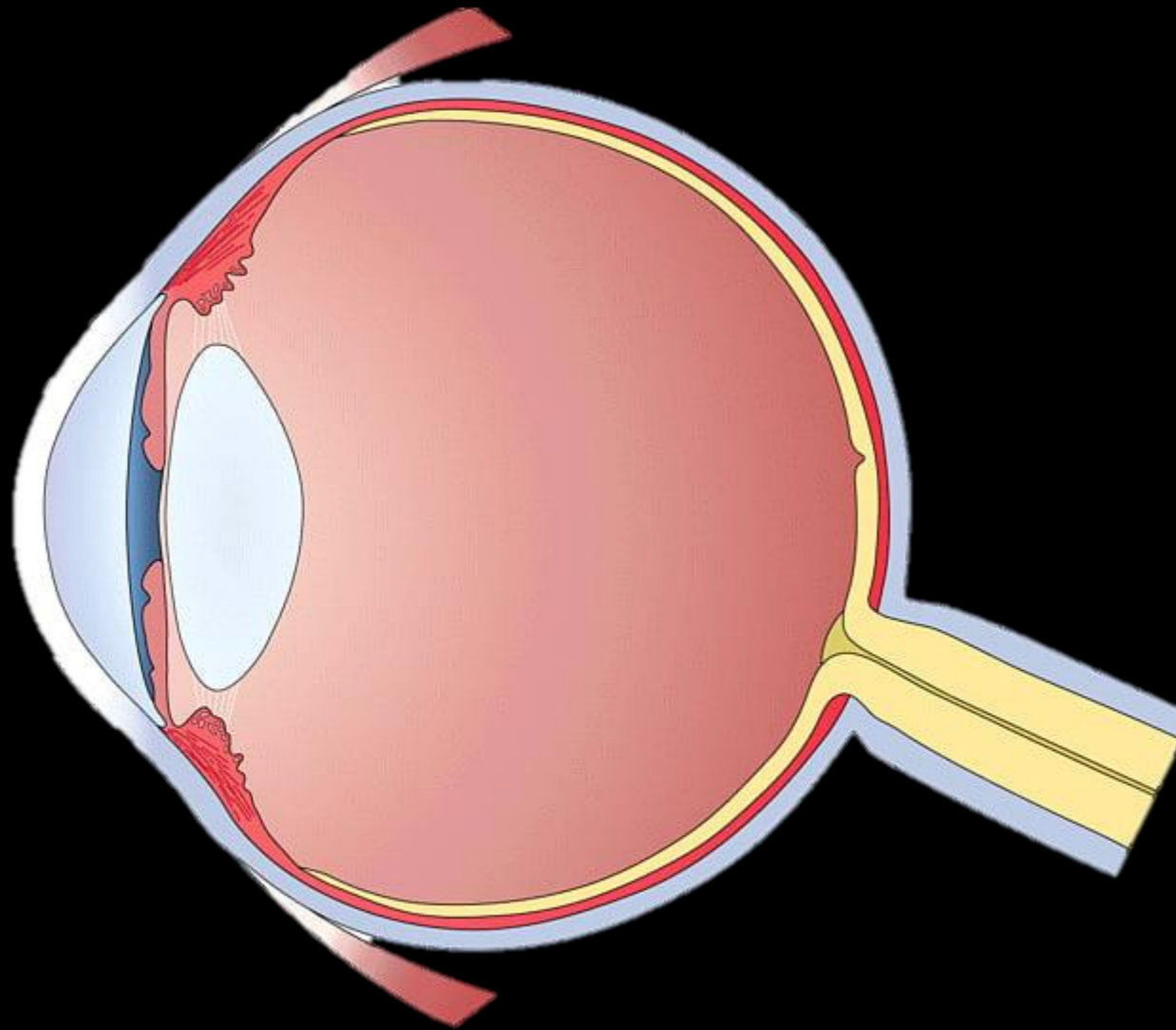


# Gap in knowledge

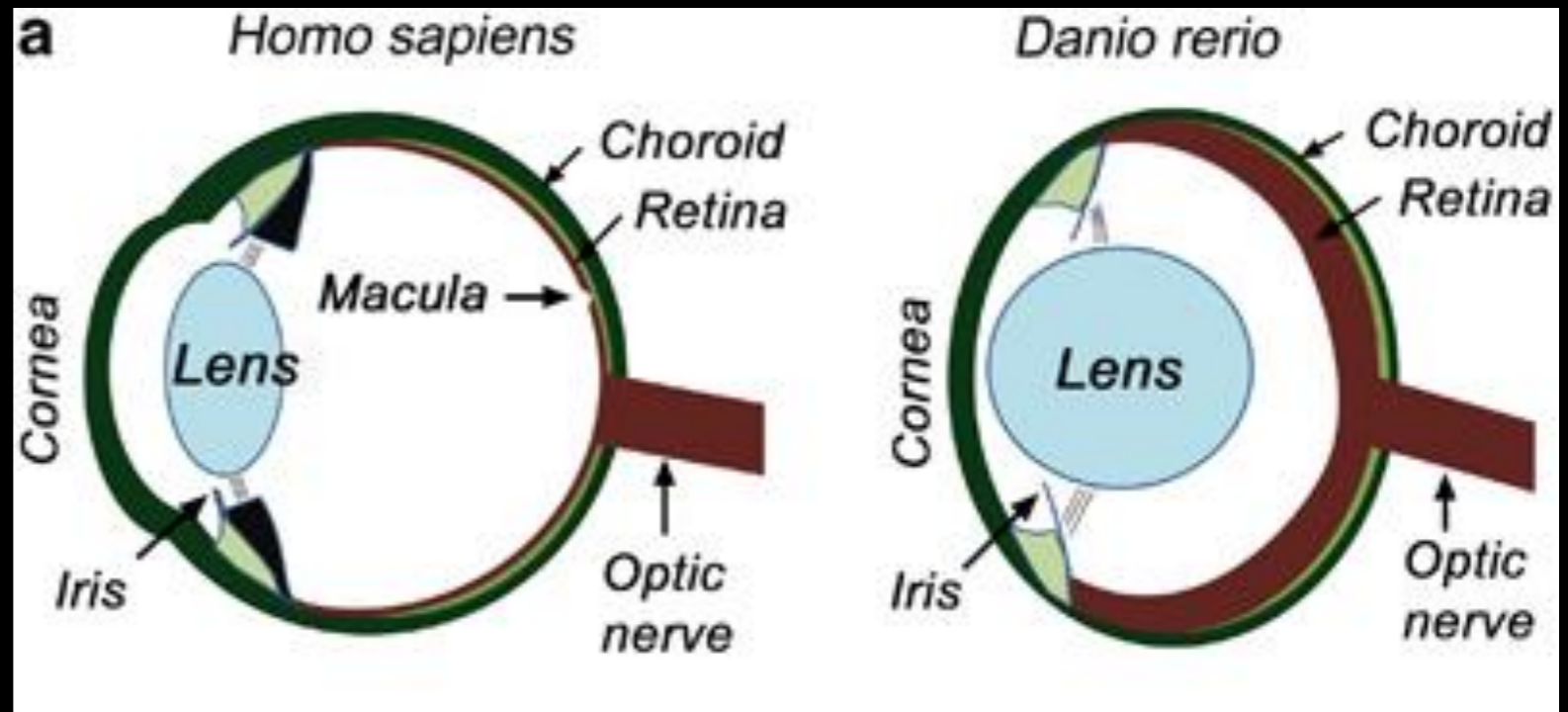
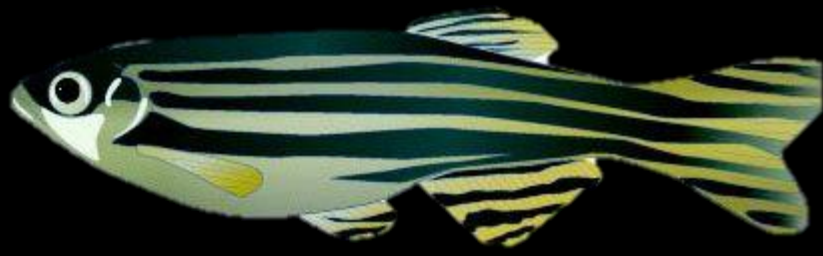


Lipofuscin buildup

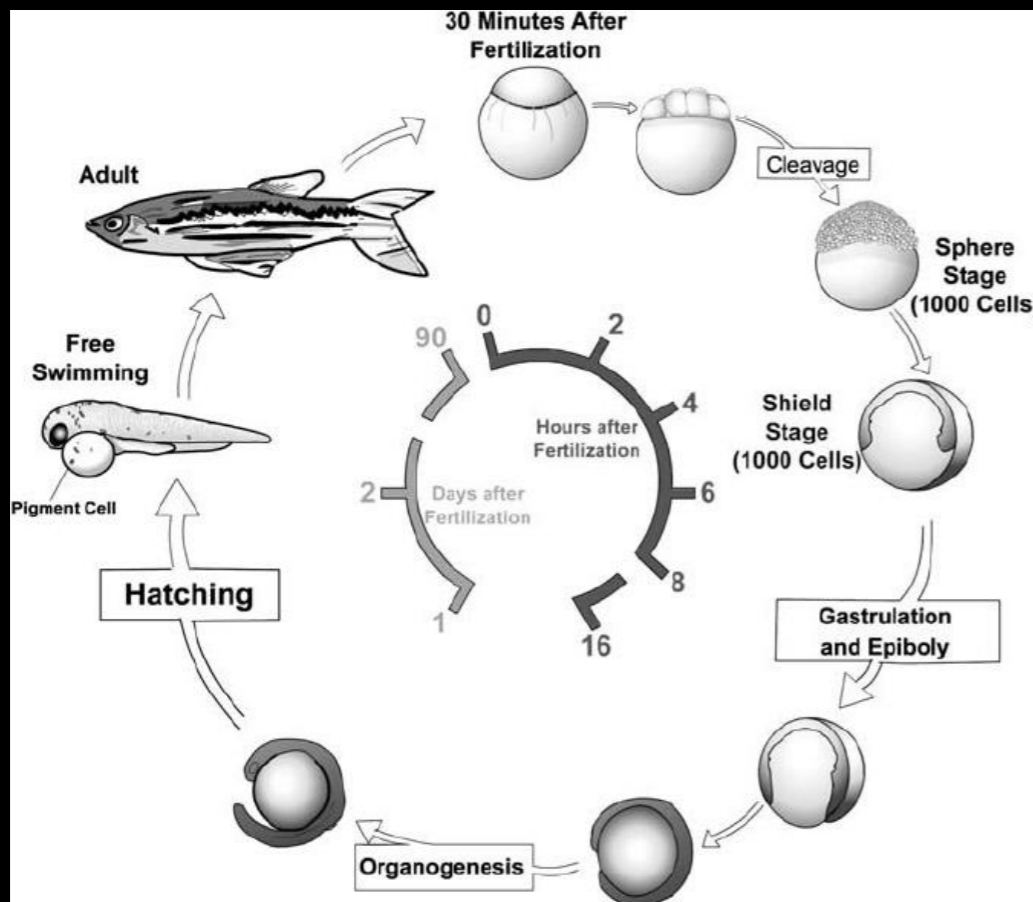
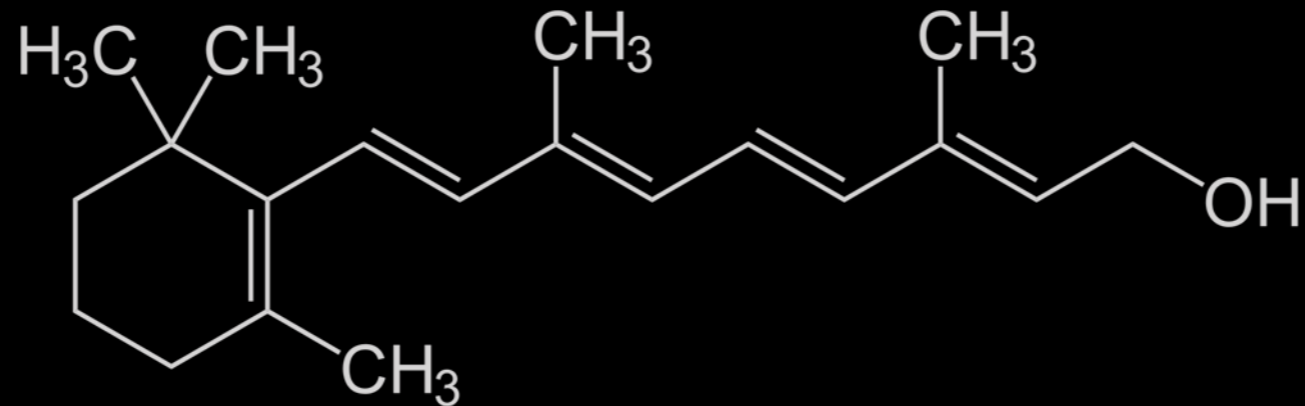
What model organism is best to study ABCA4 and Stargardt disease?



# Why is Zebrafish a great model species for ABCA4?



# Primary goal: Determine function of ABCA4 in retinol regulation



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

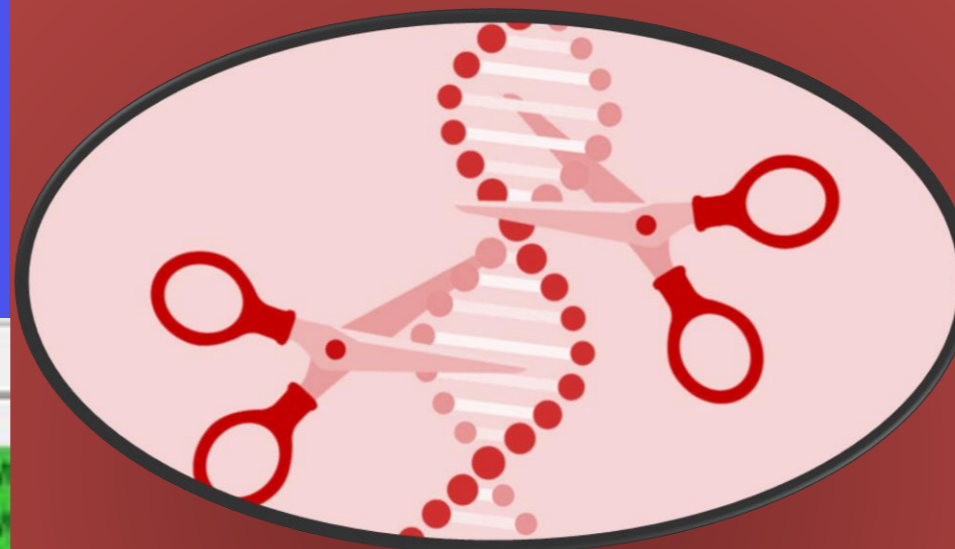
## Align sequences



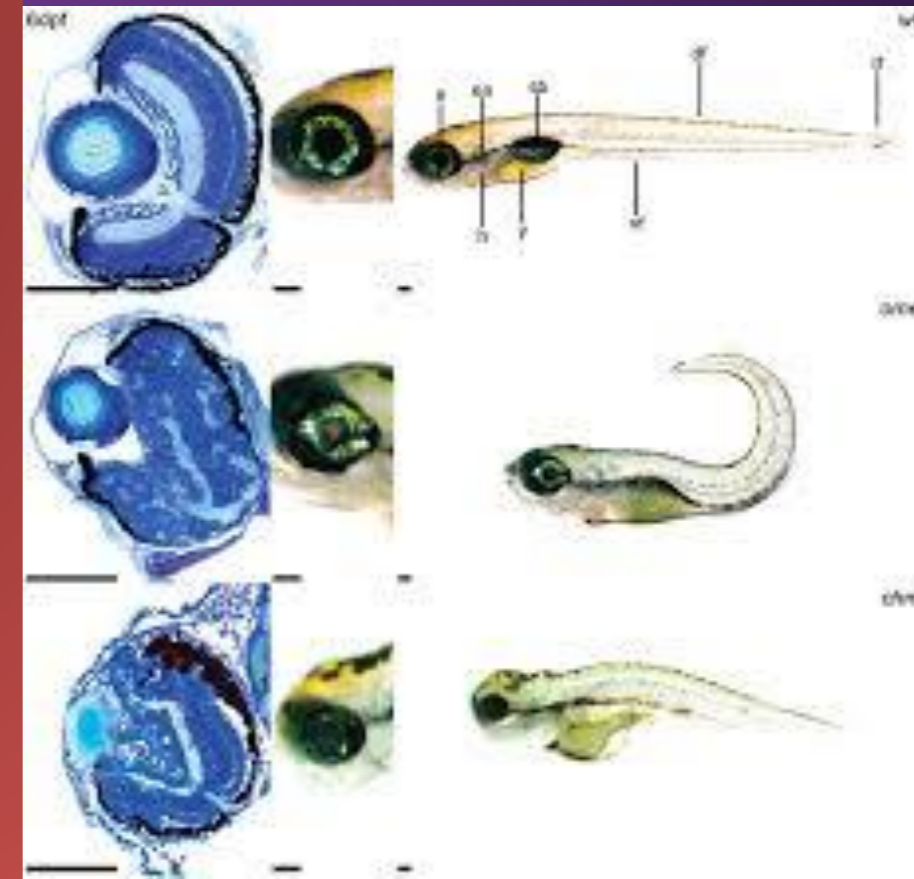
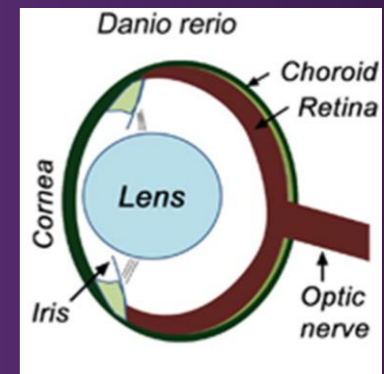
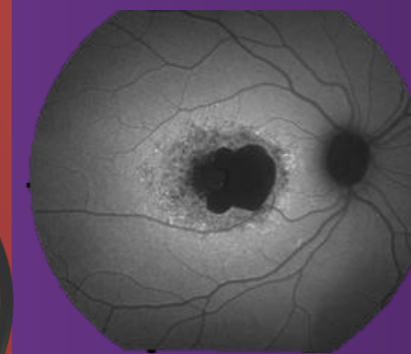
Protein Sequences

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

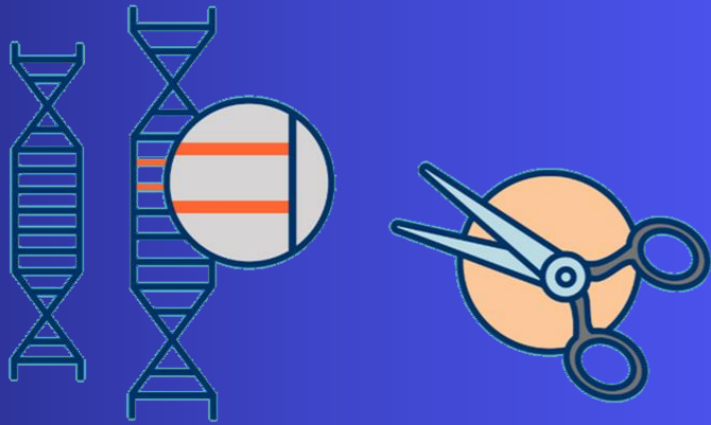


## Screen phenotypes

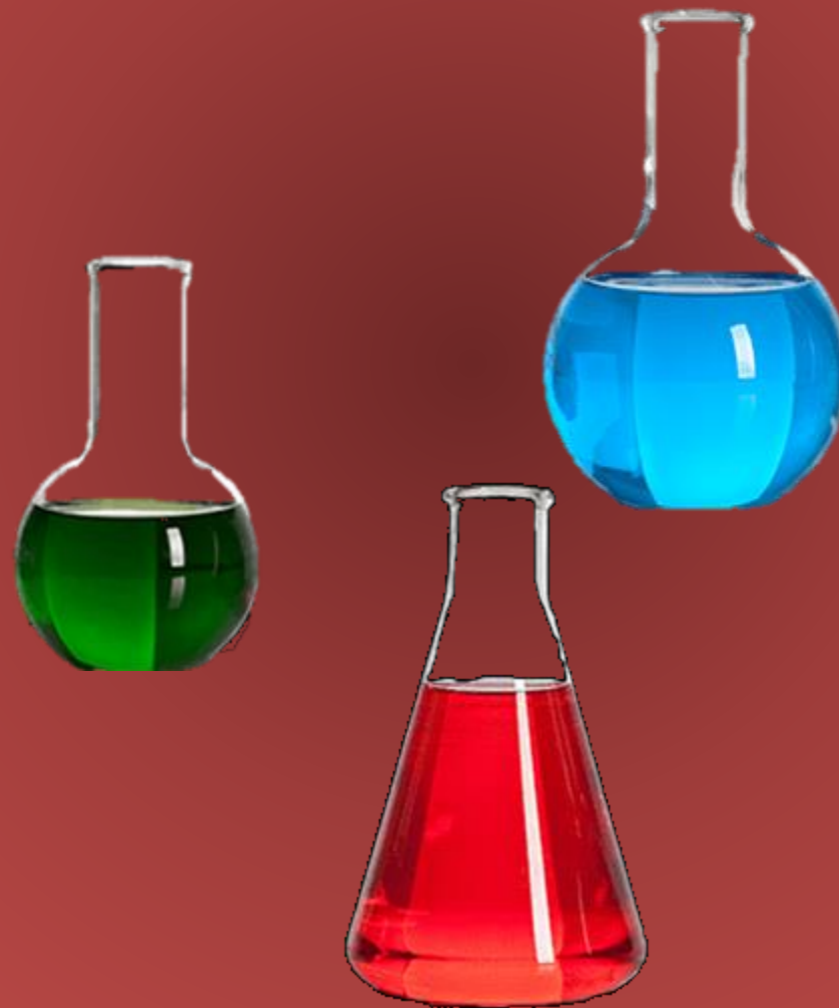


# Aim 2: Determine modulation of retinol metabolism

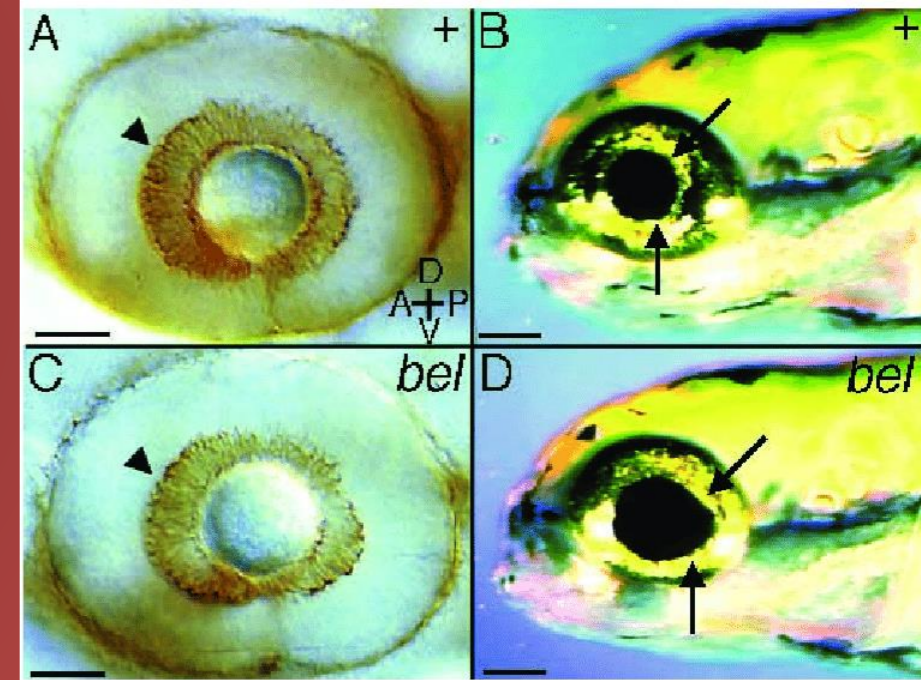
Develop mutant and WT ABCA4 populations



Perform chemical screen

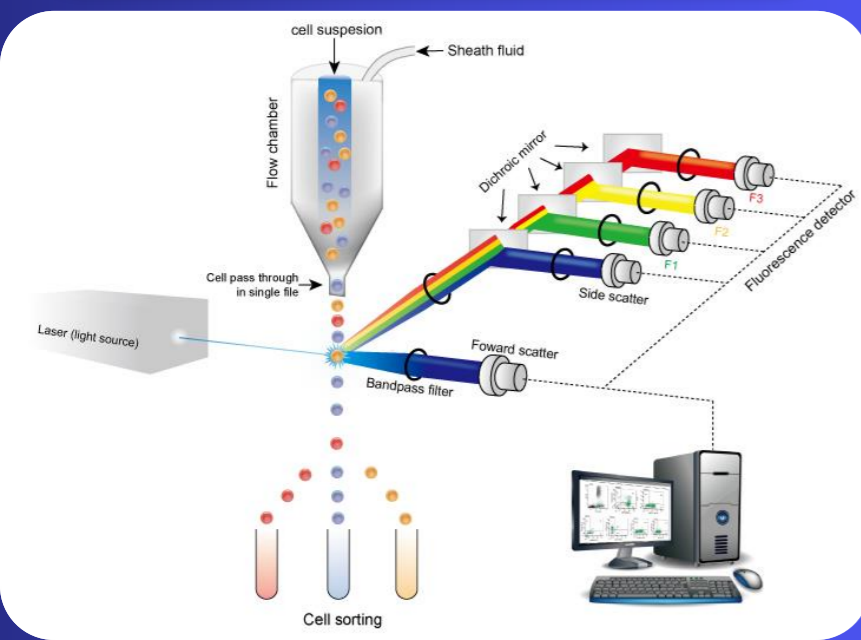
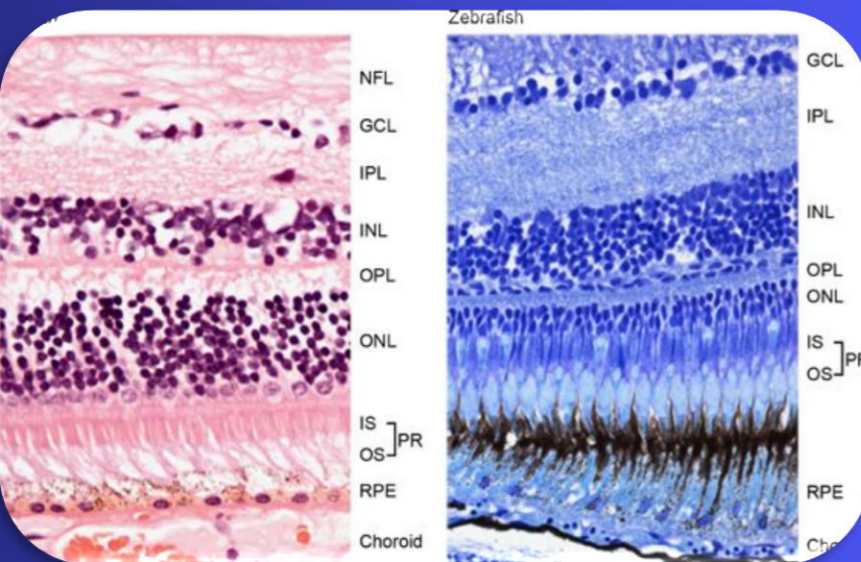


Analyze resulting phenotypes



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

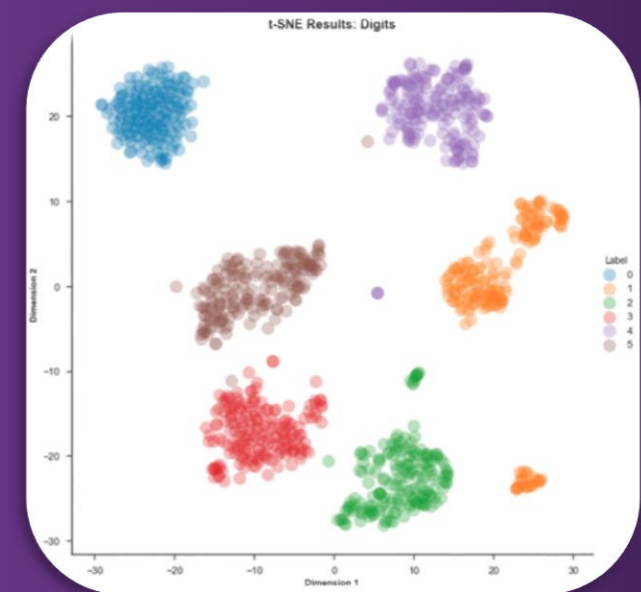
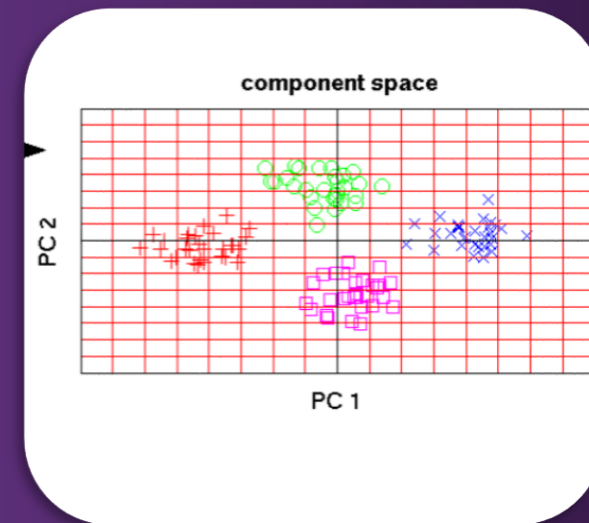
## Isolate cells



## Single-Cell RNA Sequencing



## Dimensional reduction and analysis

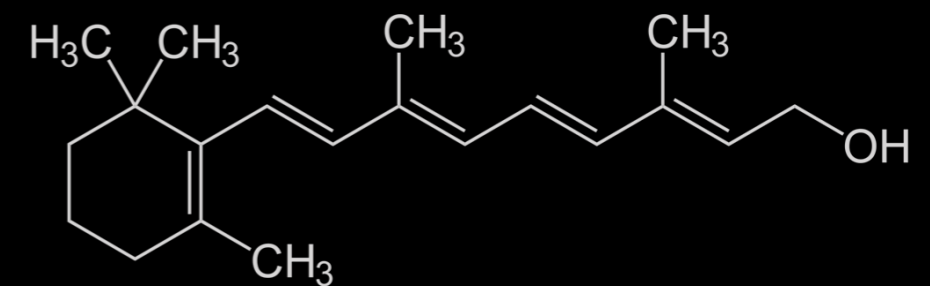


# Conclusion

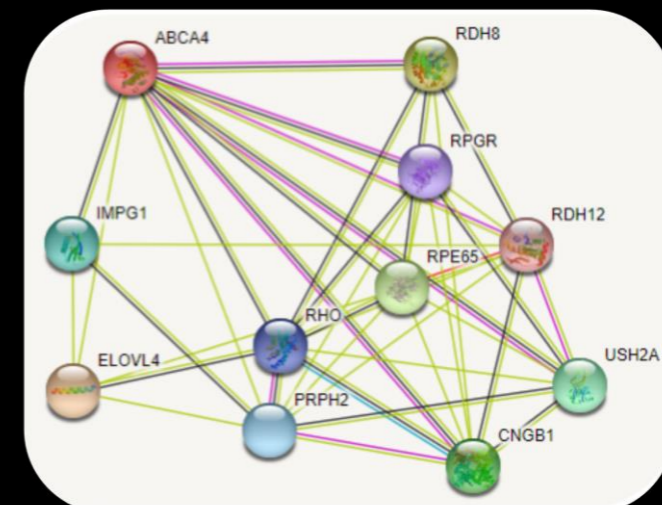
Alignment of sequences and determination of mutant Zebrafish phenotypes may assist with understanding of similar disease phenotypes in humans.

Protein Sequences	
Species/Abbrv	
1. ENSOARP00000018379	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. 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. ENSDIARP000000123162	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. ENSOCUP0000000845	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 RNA expression of single-cell types in the retina can further the understanding of ABCA4 expression and retinol metabolism





# Future Directions



Questions?

# Image References

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