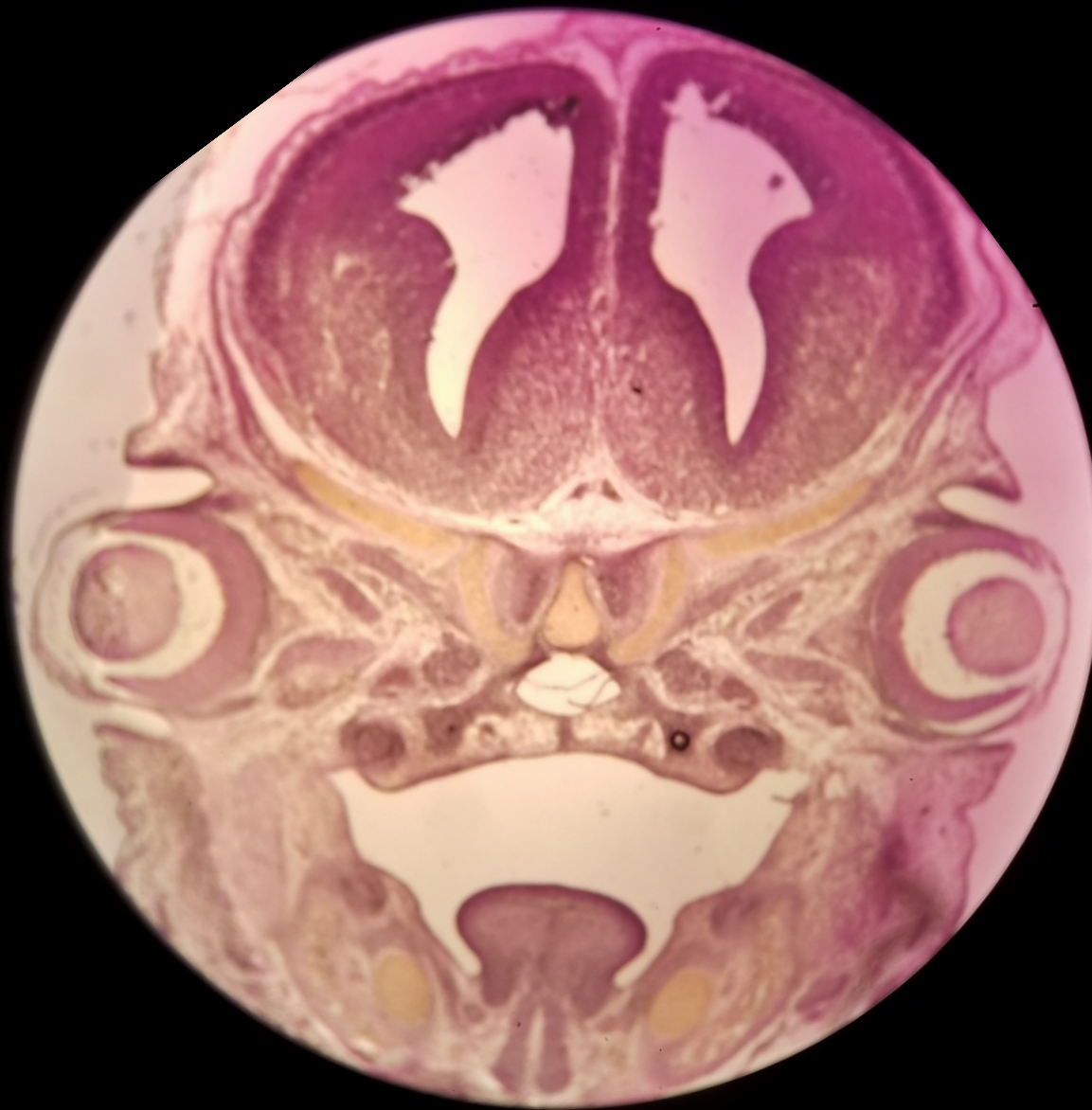




Retina

Desenvolvimento & Regeneração

Aula: Desenvolvimento e Regeneração da Retina

Roteiro

Parte 1: Revisão sobre a formação embrionária do olho

Parte 2: Diferenciação celular e Fatores de determinação

Parte 3: Desenvolvimento da Retina

Parte 4: Regeneração retiniana

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Departamento de Biologia Celular e do Desenvolvimento
Instituto de Ciências Biomédicas
carol.deldebbio@usp.br

Aula: Desenvolvimento e Regeneração da Retina

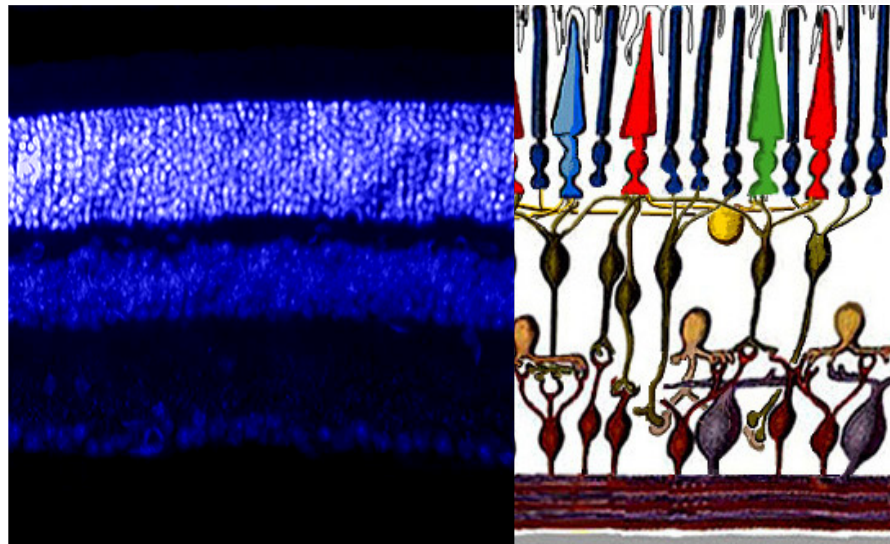
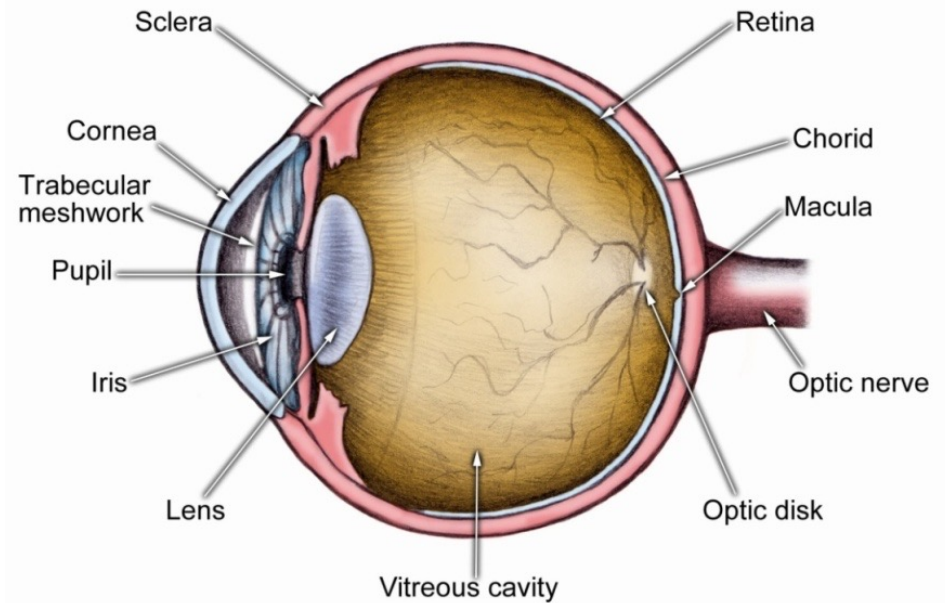
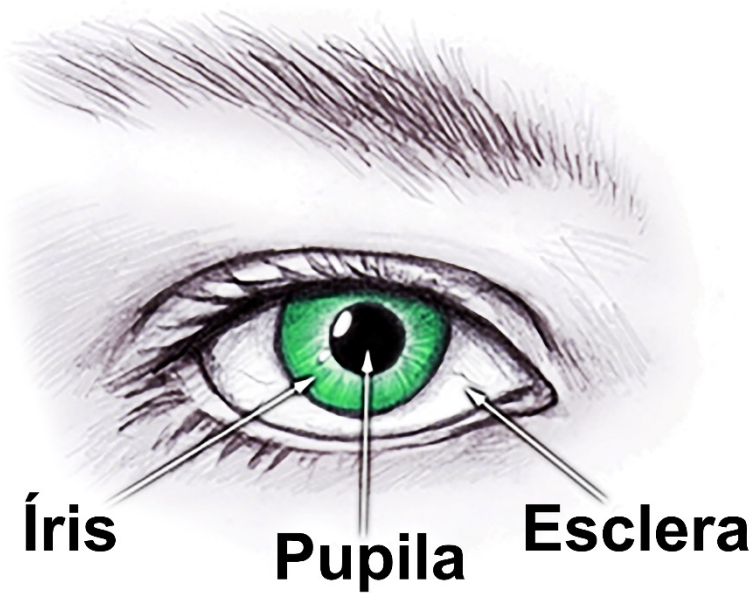
Parte 1 - Revisão sobre a formação embrionária do olho

Apesar de parecerem diferentes, os olhos de vertebrados possuem constituição bem semelhante

Desenvolvimento embrionário com etapas preservadas entre espécies



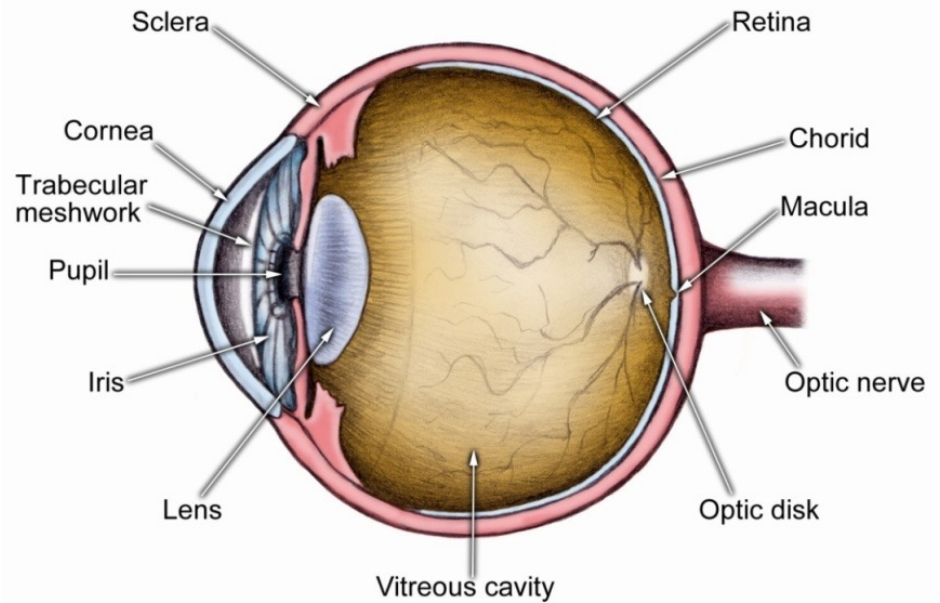
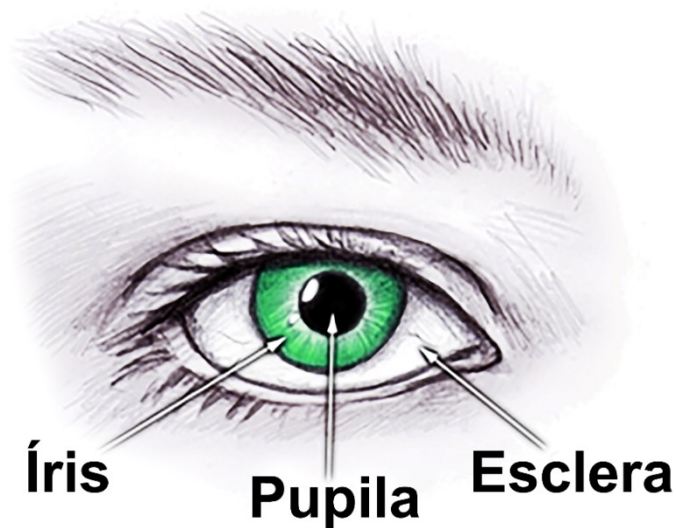
Estrutura do Olho



Fotorreceptores

**Horizontais
Bipolares
Amácrinas**

**Células Ganglionares
Nervo Óptico**



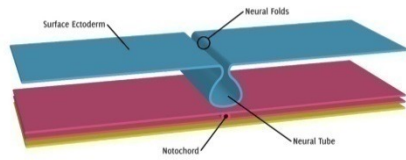
Estruturas embrionárias que originam o olho:

Cristalino e parte da córnea = **ectoderma** (não neural)

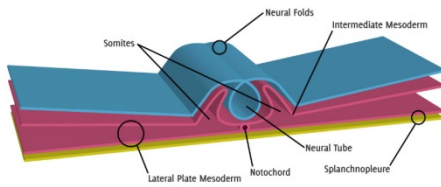
Retina e Epitélio Pigmentado = **Neuroectoderma**

Músculos Ciliares, Íris e Esclera = **Crista Neural**

Parte da Córnea = **Mesoderma**

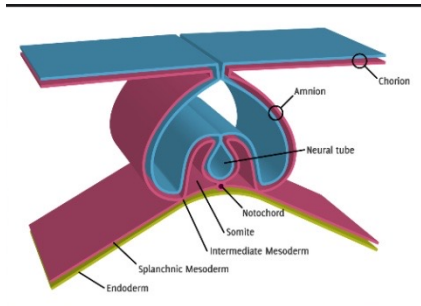


GASTRULAÇÃO

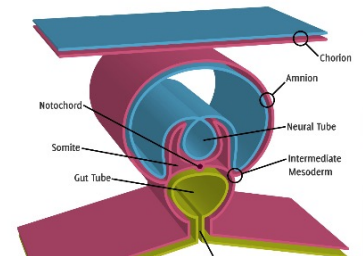


NEURULAÇÃO

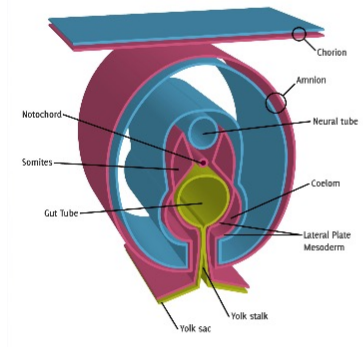
Formação da Vesícula Óptica



DOBRAMENTO DO CORPO



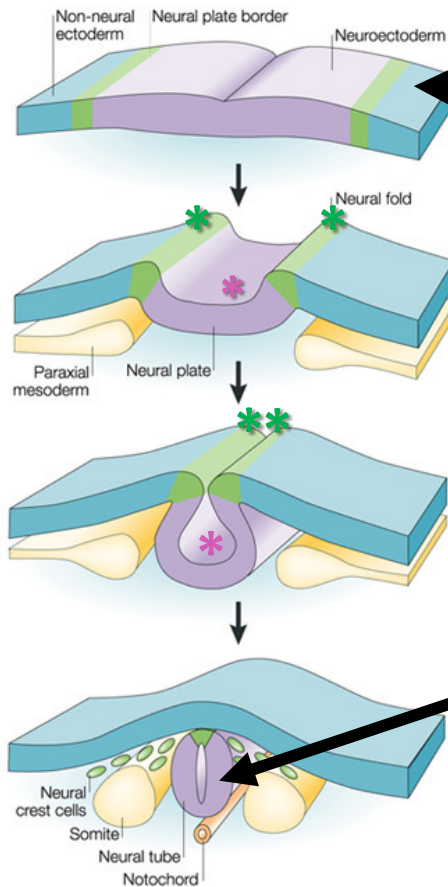
FECHAMENTO DO TUBO ENDODÉRMICO



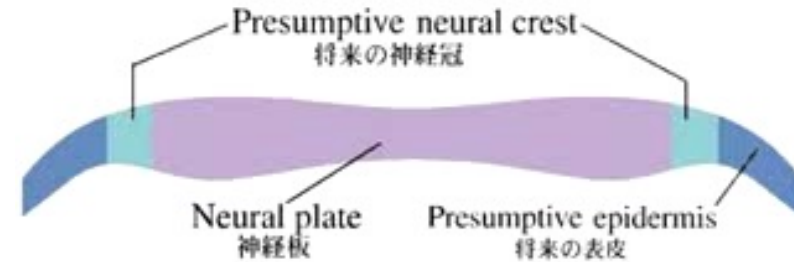
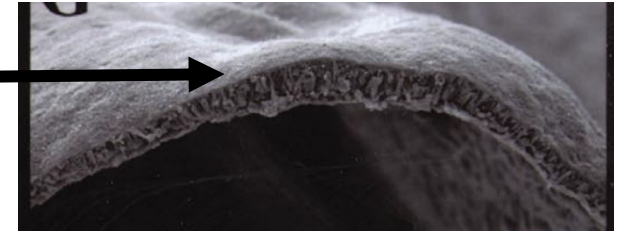
EXPANSÃO DO ÂMNION

Neurulação: formação do tubo neural (futuro SNC)

Dobramento da **placa neural** e Encontro das **pregas neurais**

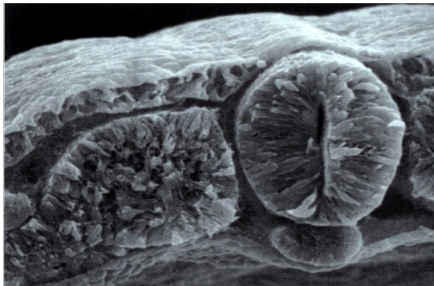


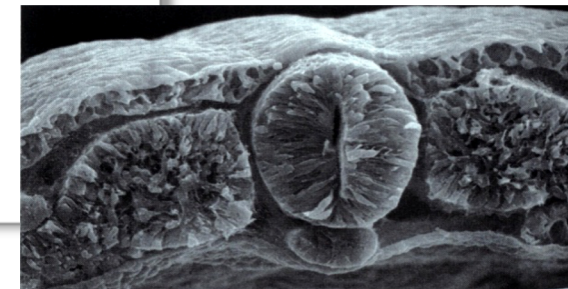
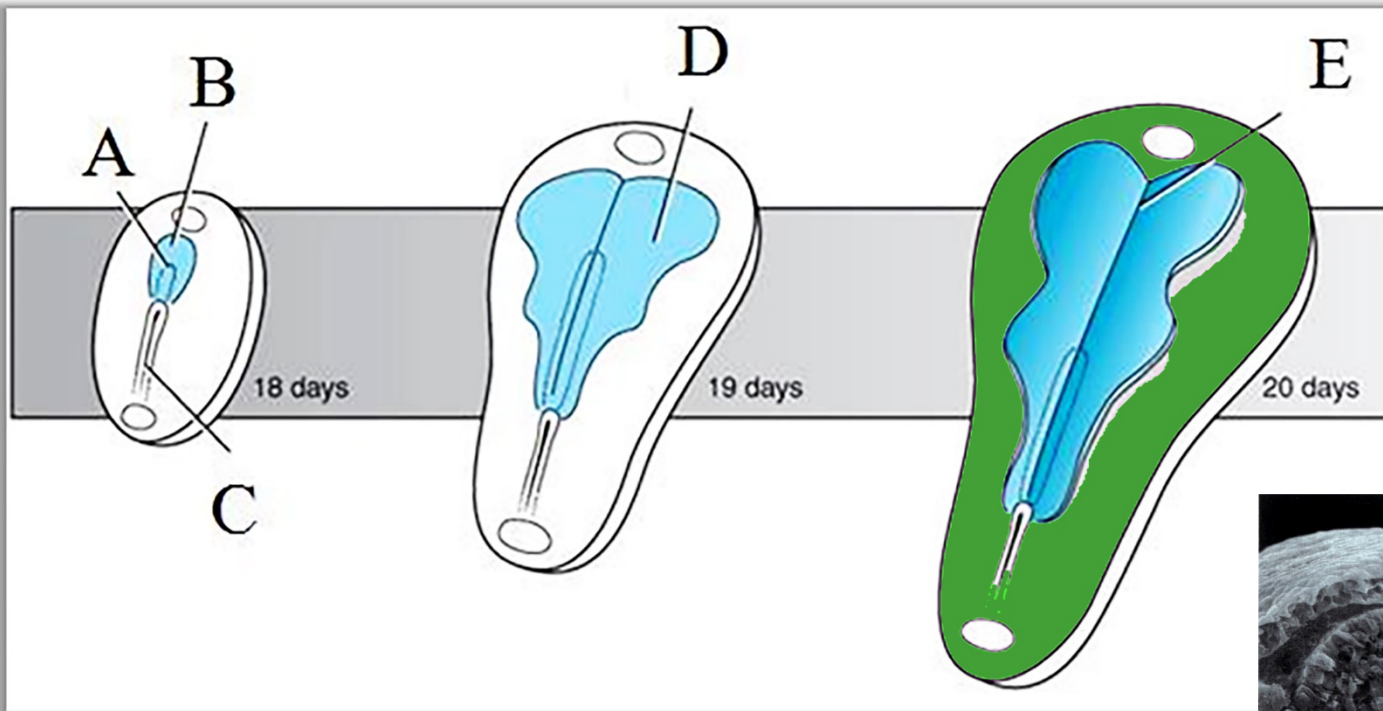
ectoderme



<https://www.youtube.com/watch?v=NkTy-sNpnI8>

Tubo neural





ECTODERMA

NEUROECTODERMA

Tubo Neural

Cérebro, medula,
retina, neuro-
hipófise

Crista Neural

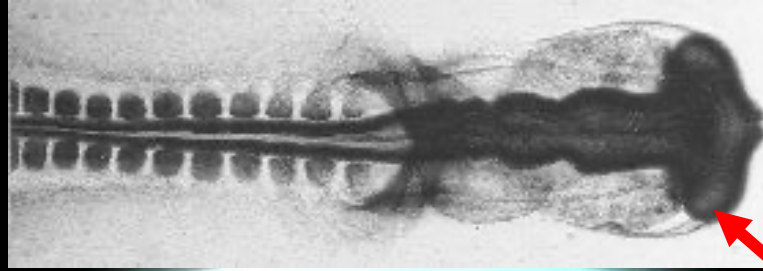
Gânglios e nervos cranianos e
sensitivos, células pigmentares,
cartilagem dos arcos branquiais,
mesênquima da cabeça

ECTODERMA SUPERFICIAL

Epiderme, pelos, unhas, esmalte
dentário, glândulas cutâneas e mamárias,
lobo anterior da hipófise, orelha interna,
crystalino



<https://youtu.be/00xr4kzYZn4>



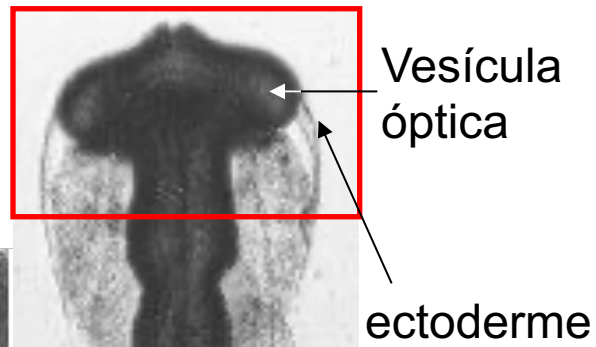
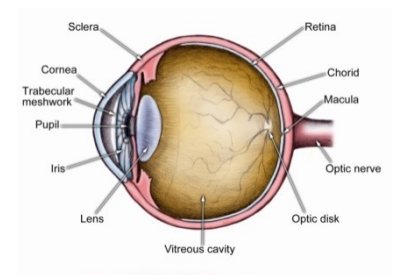
Vesícula Óptica



O olho se origina no Prosencéfalo → Telencéfalo



O olho se desenvolve a partir da **vesícula óptica** (forma a RETINA) e do ectoderme sobrejacente (forma o CRISTALINO)



Dobras neurais

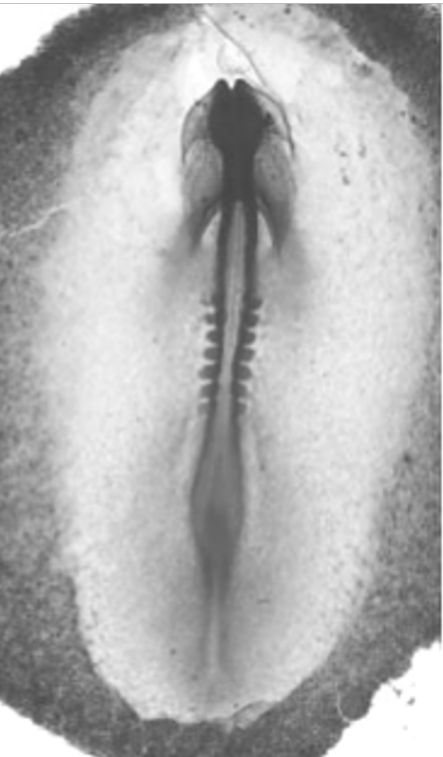
Tubo neural



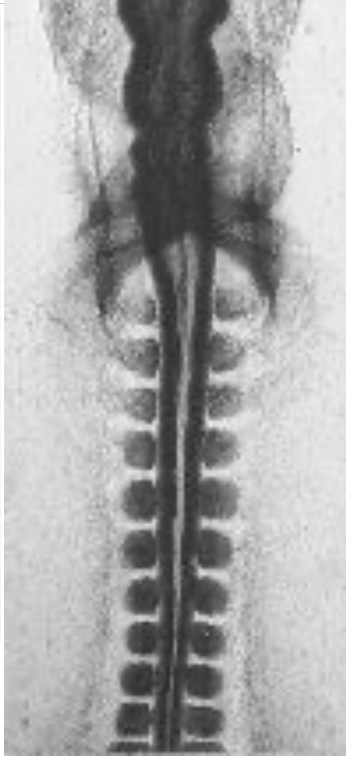
26h



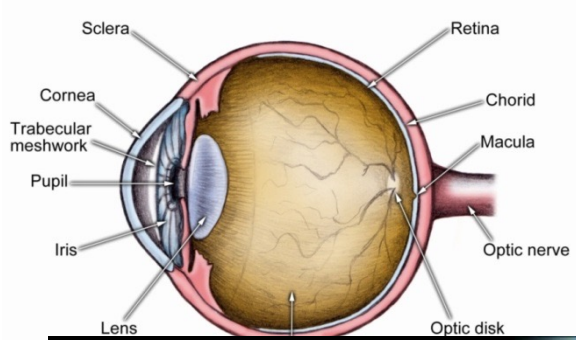
30h



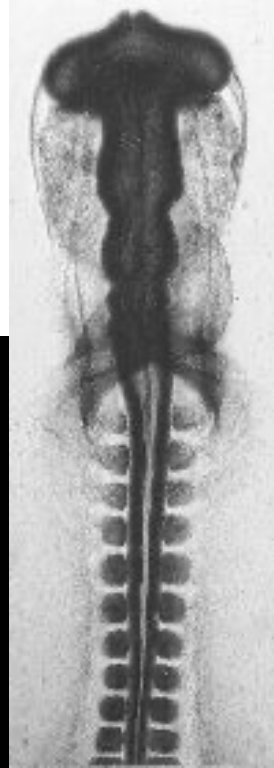
33h
(7 somitos)



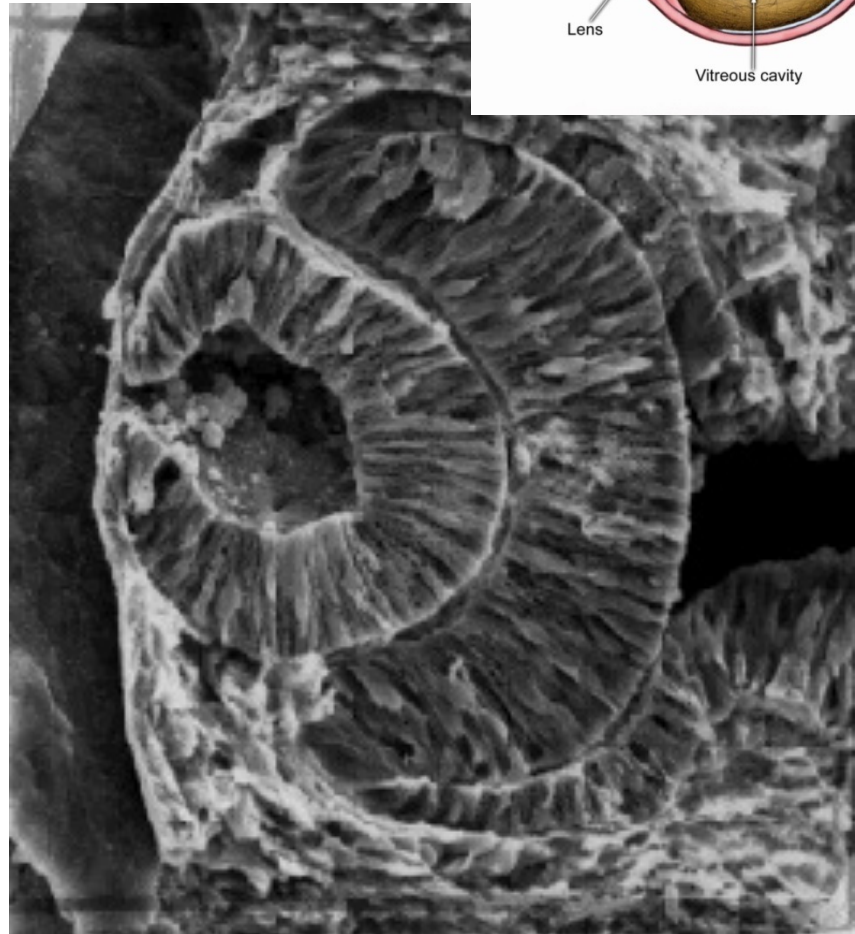
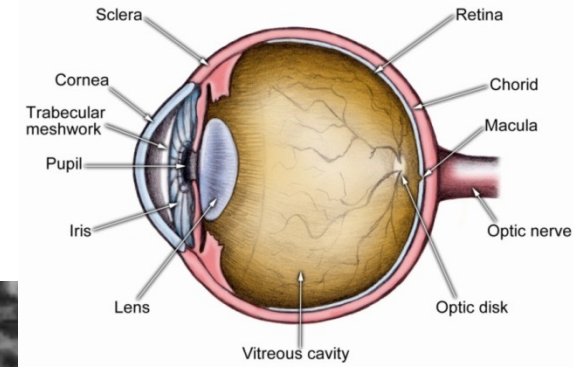
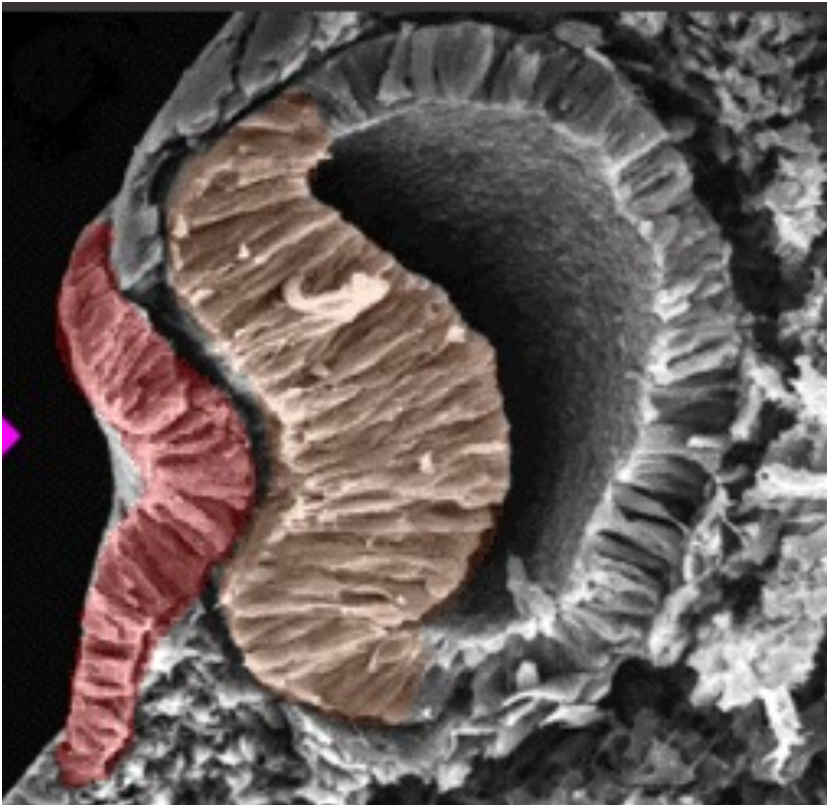
36h
(10 somitos)



Crescimento da vesícula óptica
 Invaginação vesícula – cálice óptico
 Invaginação do ectoderma - cristalino

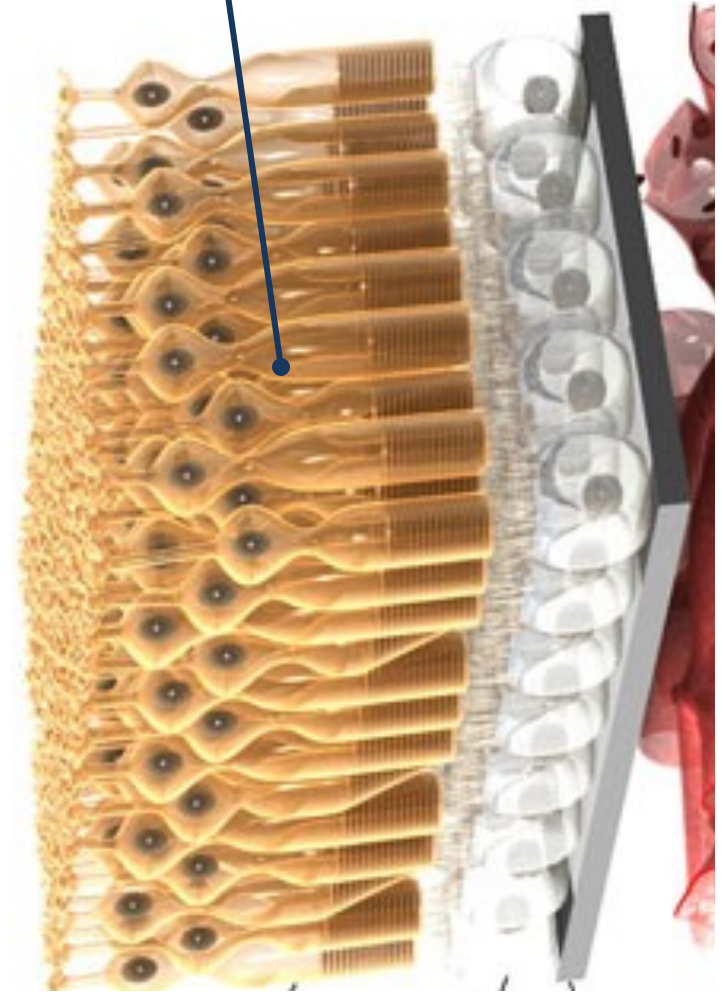
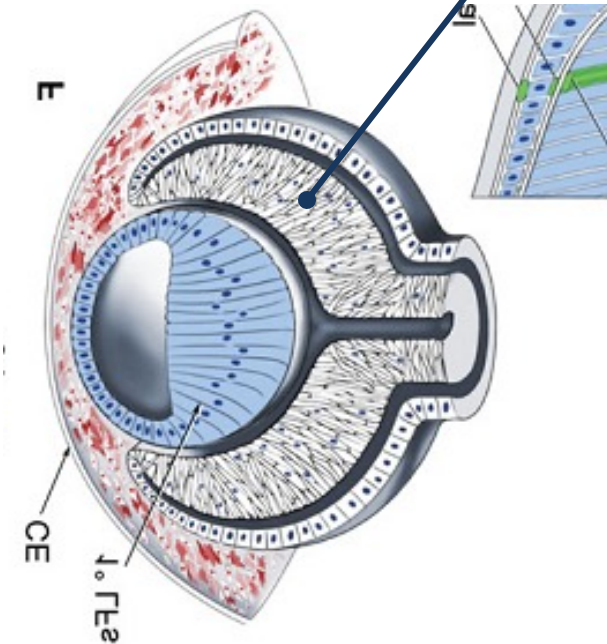
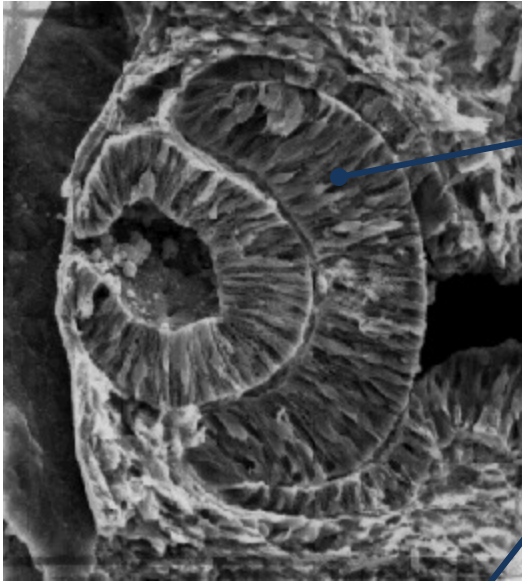


O olho se desenvolve a partir da vesícula óptica (forma a RETINA) e do ectoderme sobrejacente (forma o CRISTALINO)



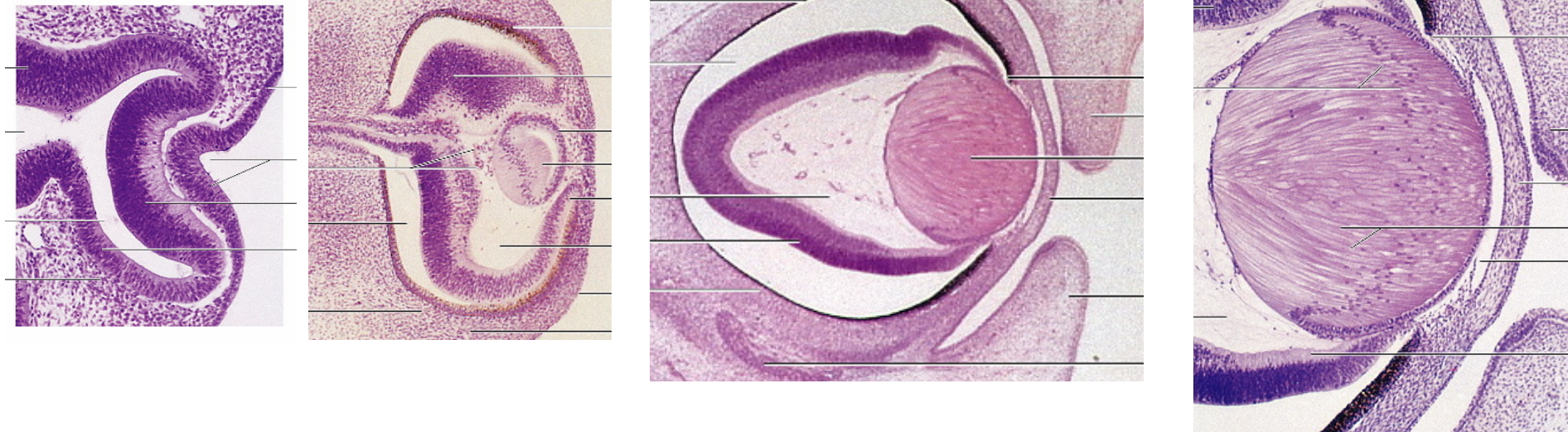
A camada interna do cálice óptico forma a

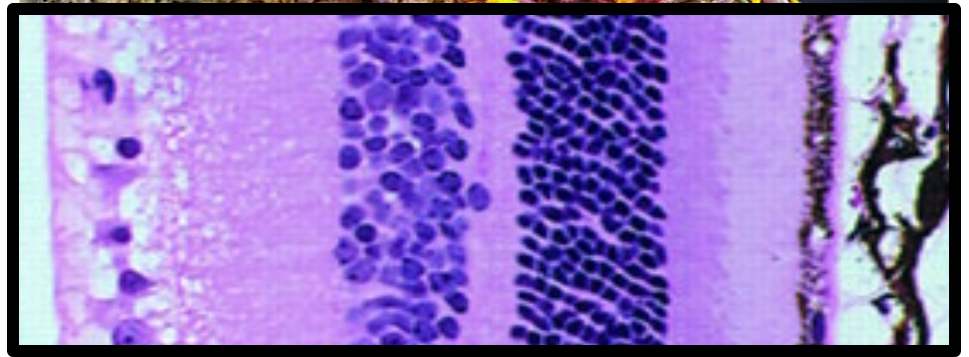
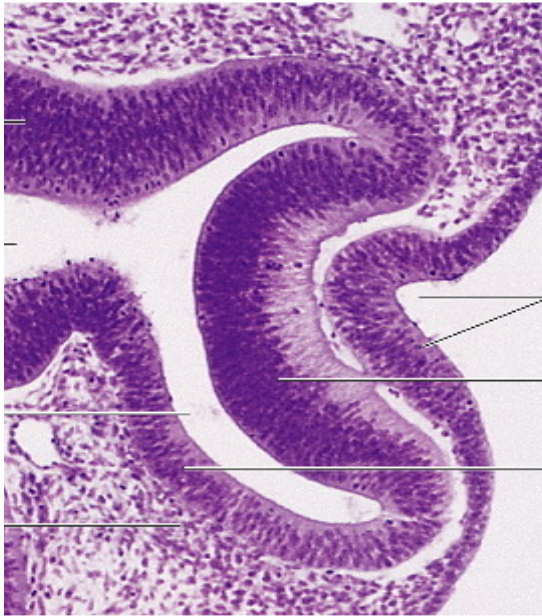
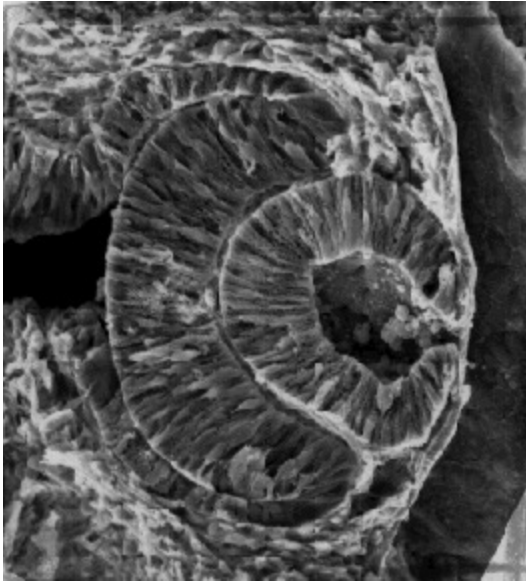
RETINA



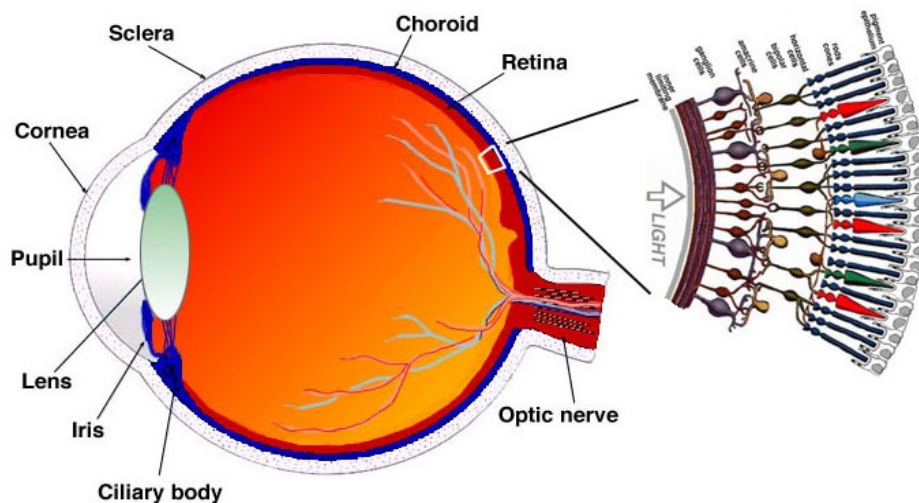
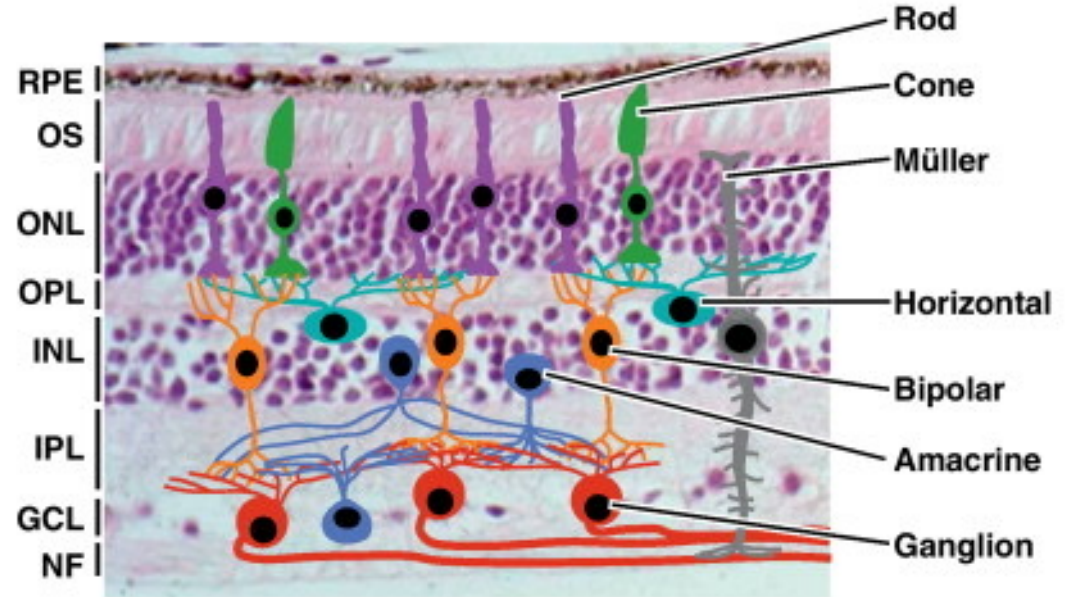
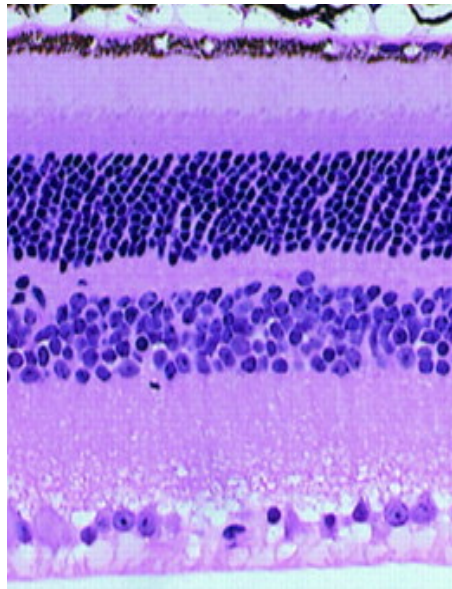
O CORPO CILIAR é formado pela “borda” do cálice óptico, onde o Epitélio Pigmentado Retiniano se encontra com a Retina

Compartilham os mesmos progenitores





A retina é composta por diferentes tipos de neurônios e glia



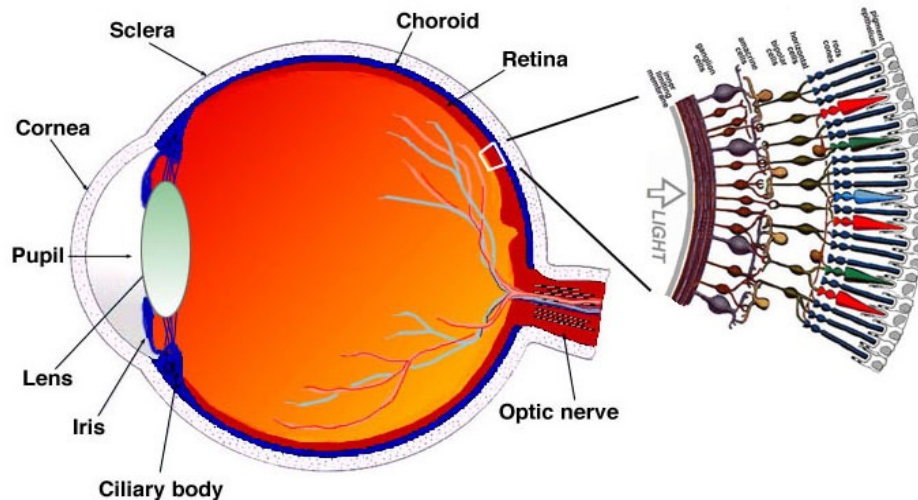
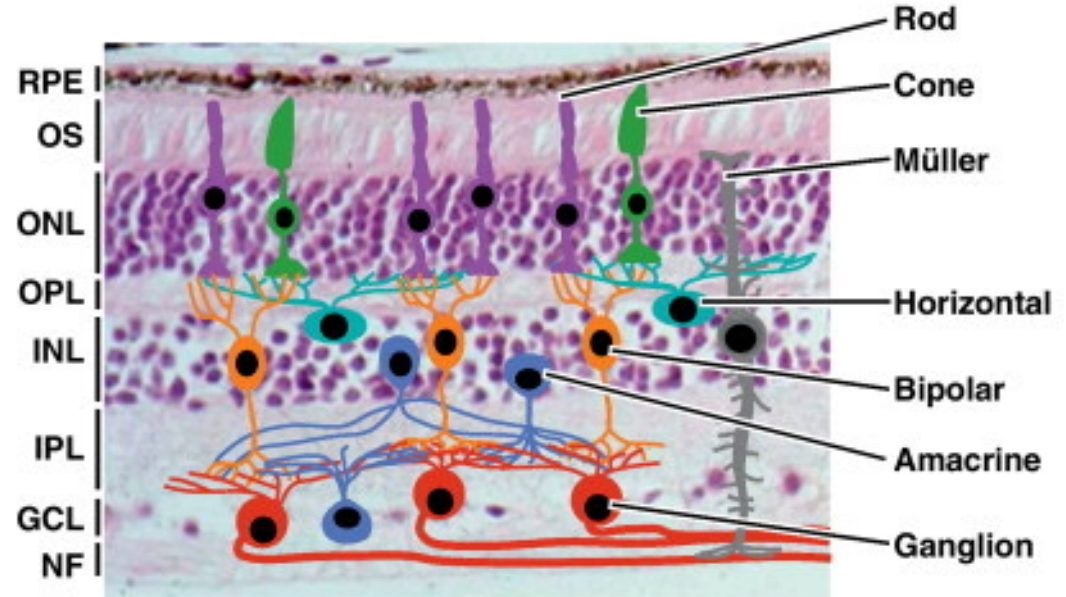
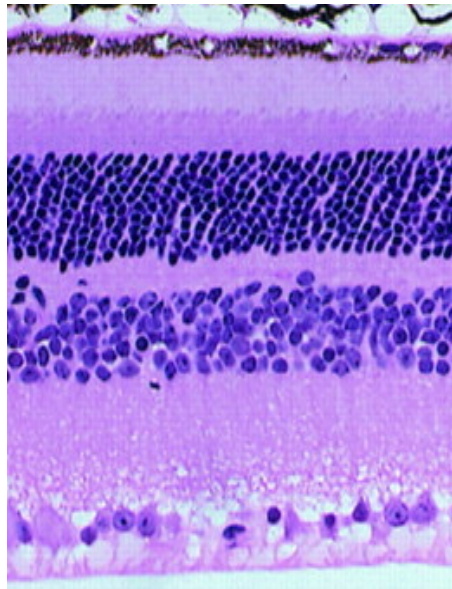
Como surgem os diferentes tipos celulares?

Determinação Celular e Fatores de Diferenciação

Aula: Desenvolvimento e Regeneração da Retina

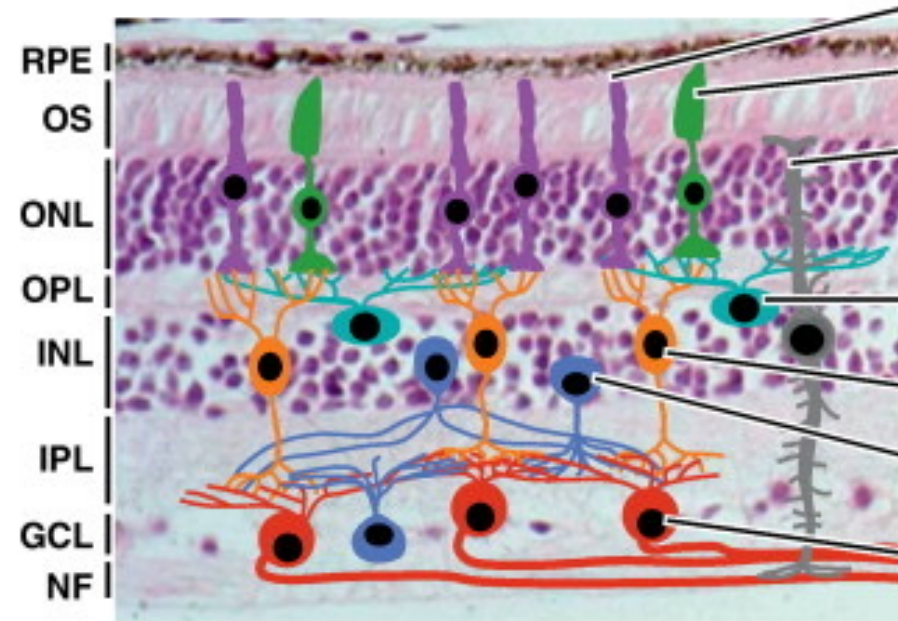
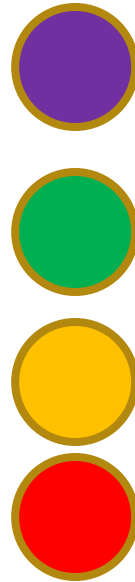
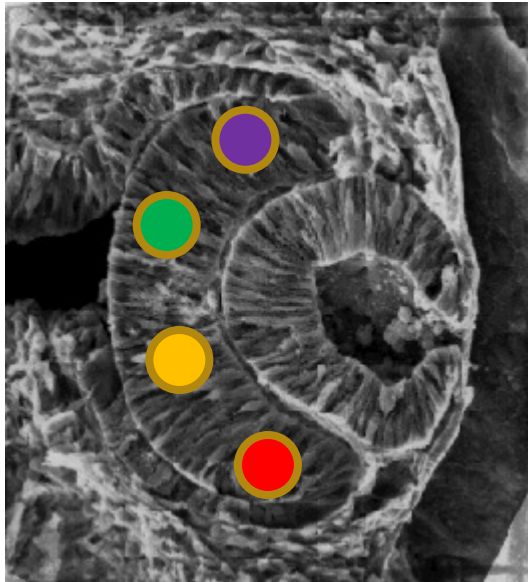
Parte 2 - Diferenciação celular e Fatores de determinação

A retina é composta por diferentes tipos de neurônios e glia

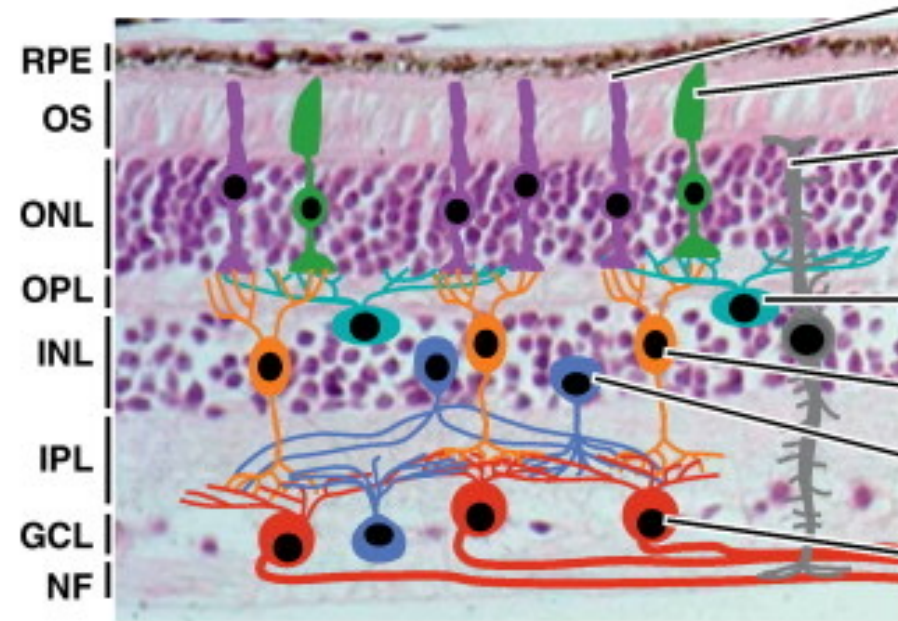
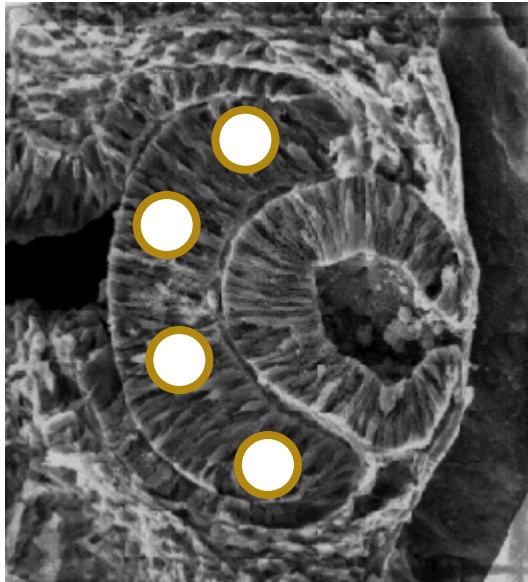


Como surgem os diferentes tipos celulares?

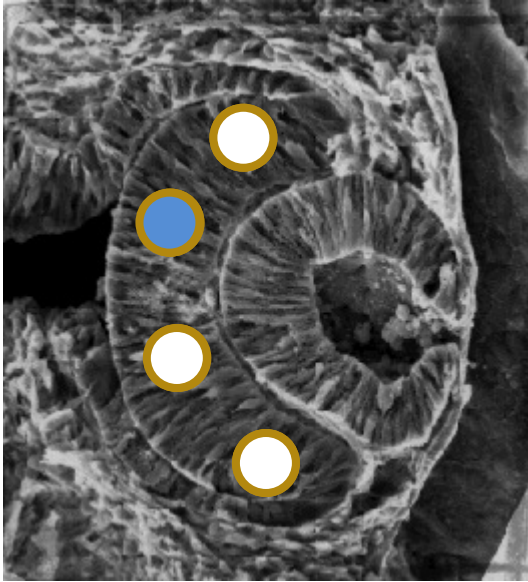
Acreditava-se que cada tipo celular era proveniente de um progenitor retiniano específico



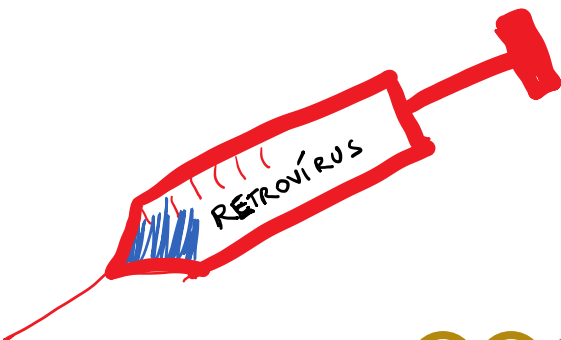
Acreditava-se que cada tipo celular era proveniente de um progenitor retiniano específico



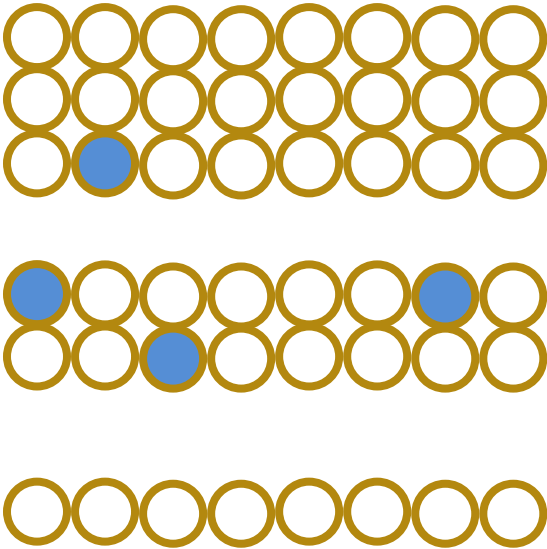
Depois, observou-se que as células partilham os mesmos progenitores



Marcador: beta-galactosidase

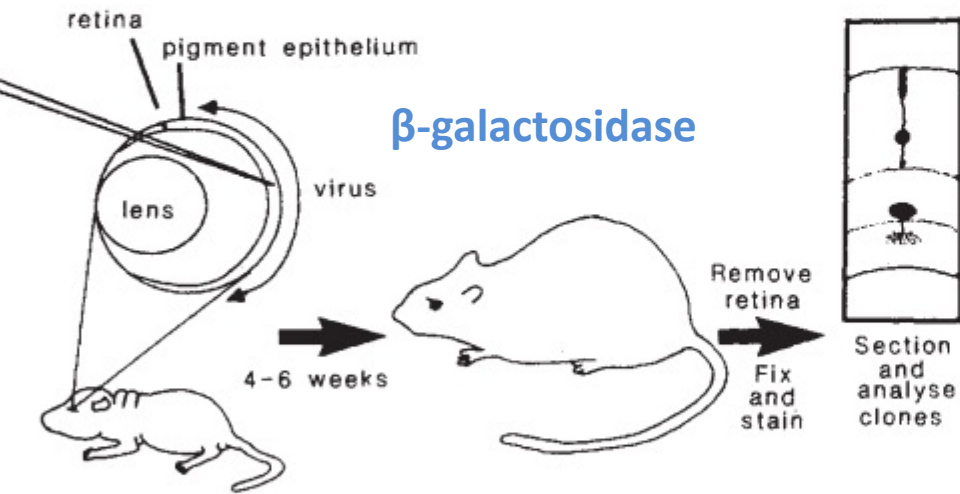


proliferação



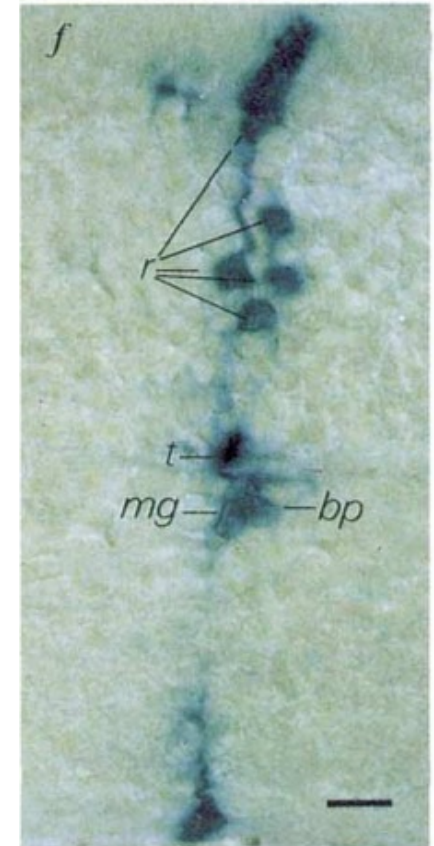
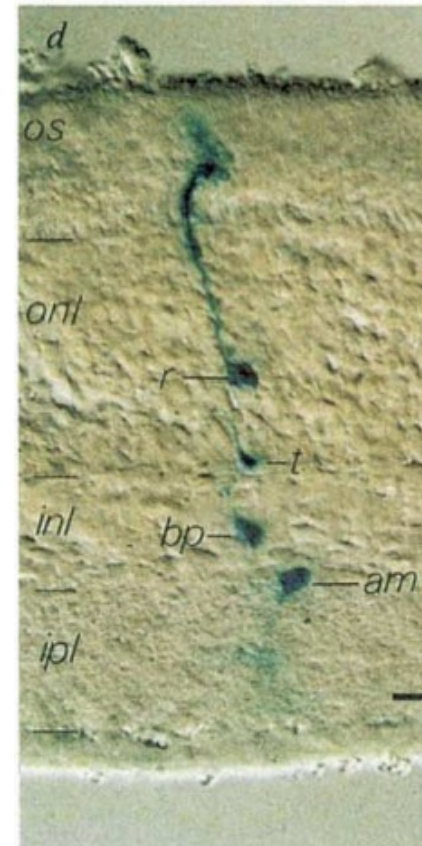
diferenciação

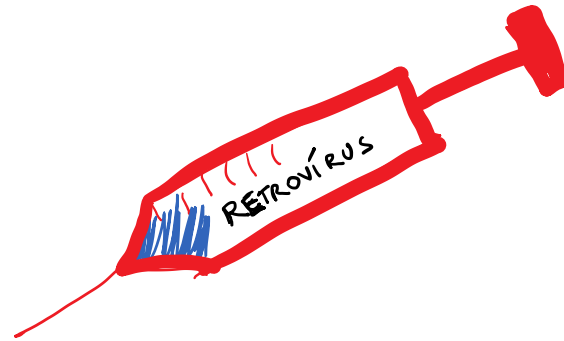
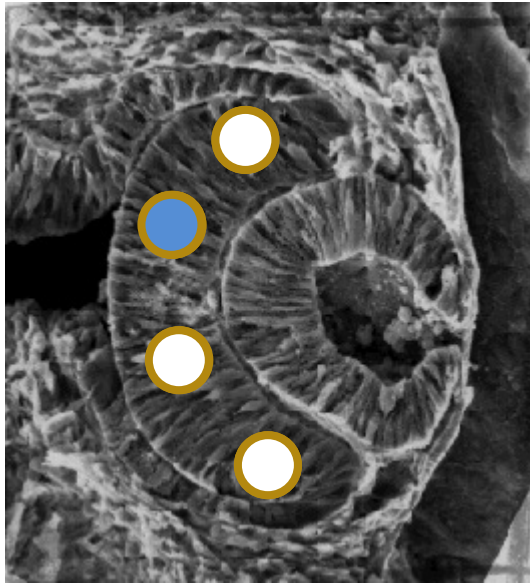
Progenitores retinianos são multipotentes



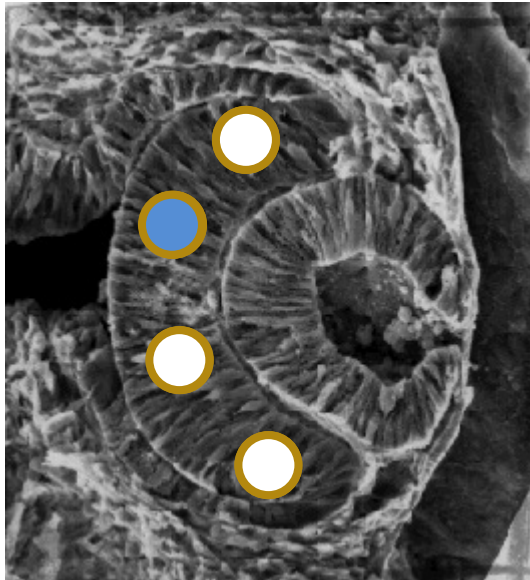
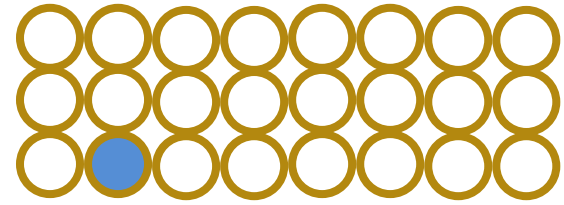
Neonato

Cones
Bastonetes
Bipolares
Amácrinas
Horizontais
Ganglionares
Müller



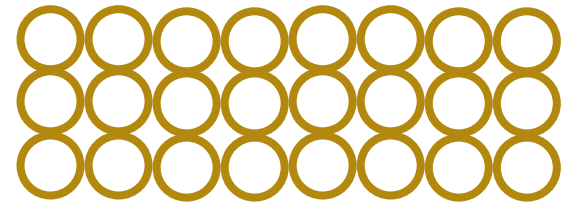


Neonato

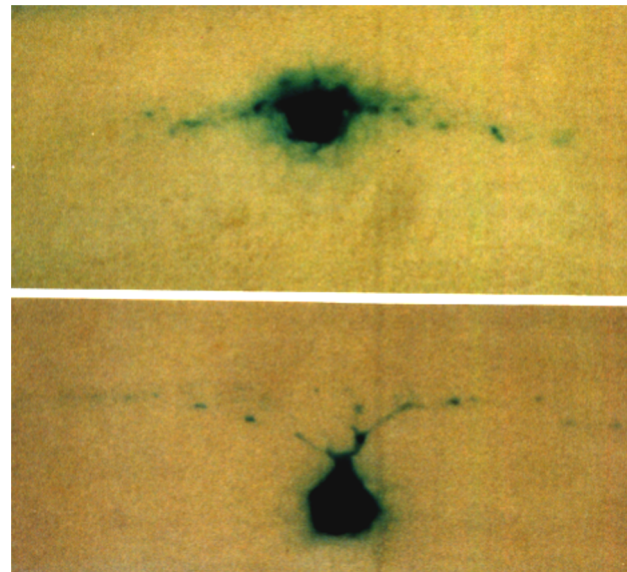
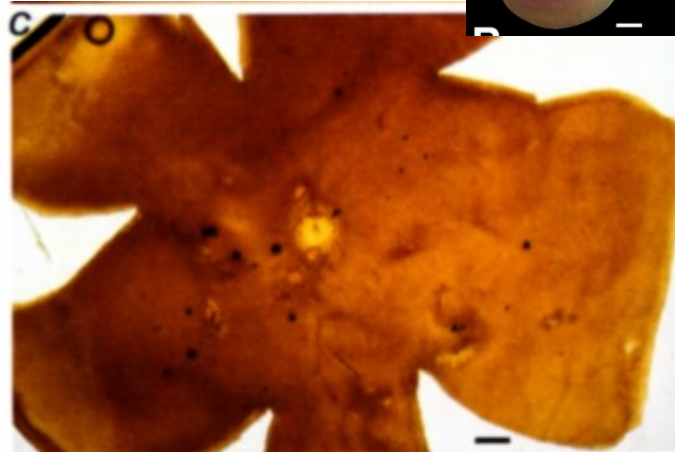
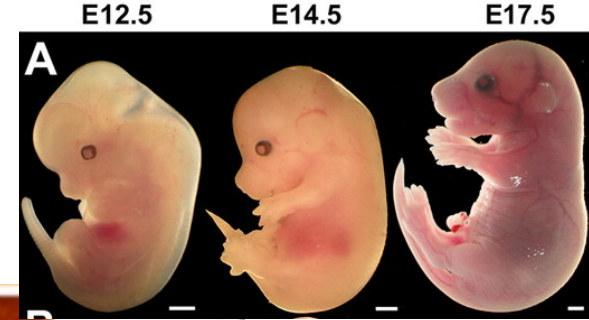
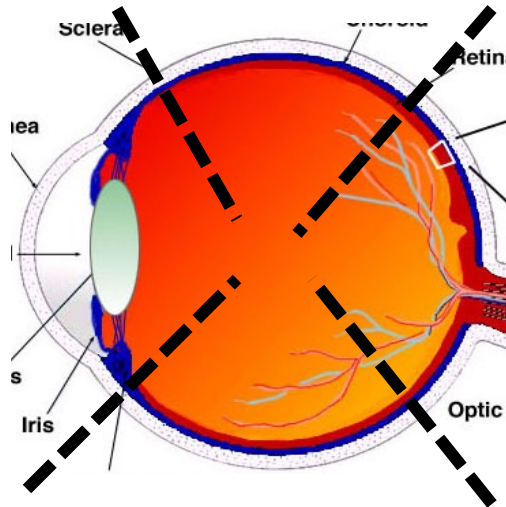


Embrião E13

Handwritten red question marks: ? ? ? ? ?

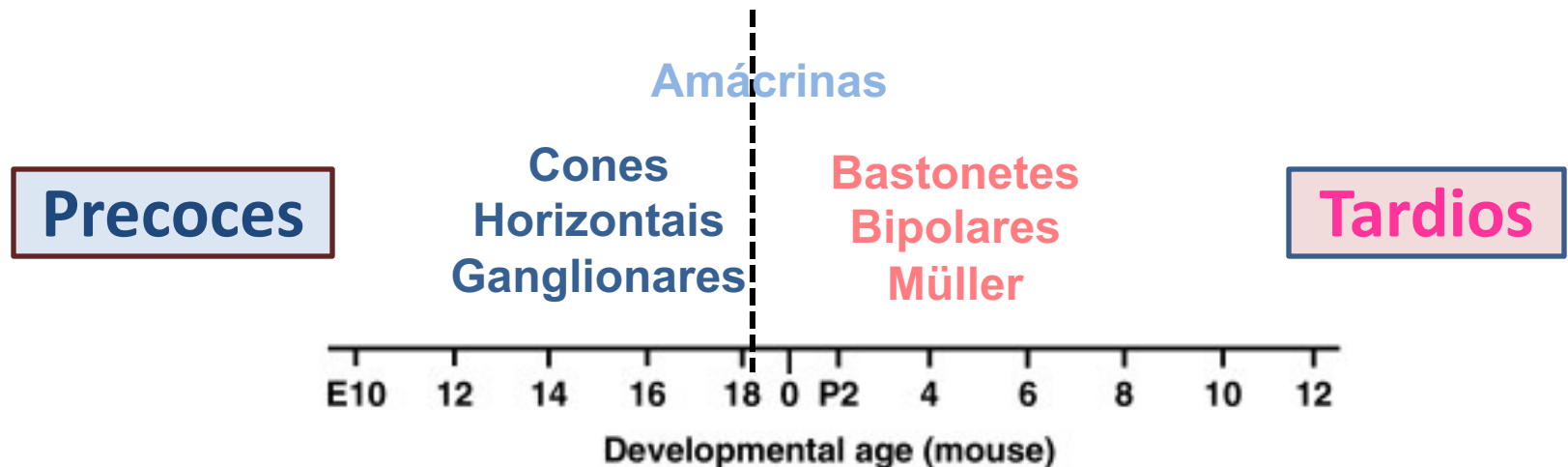
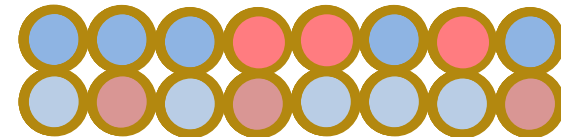
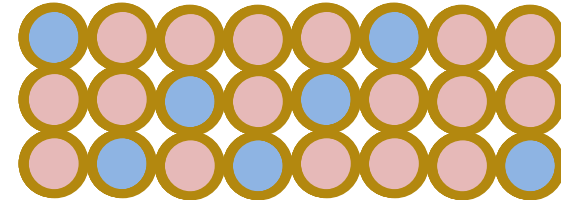
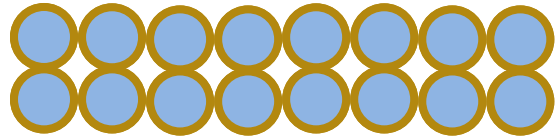
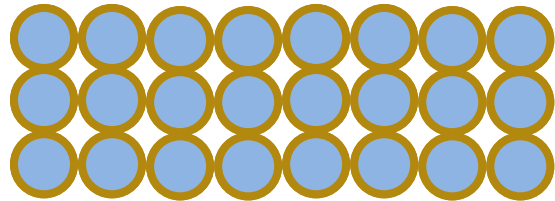


E13

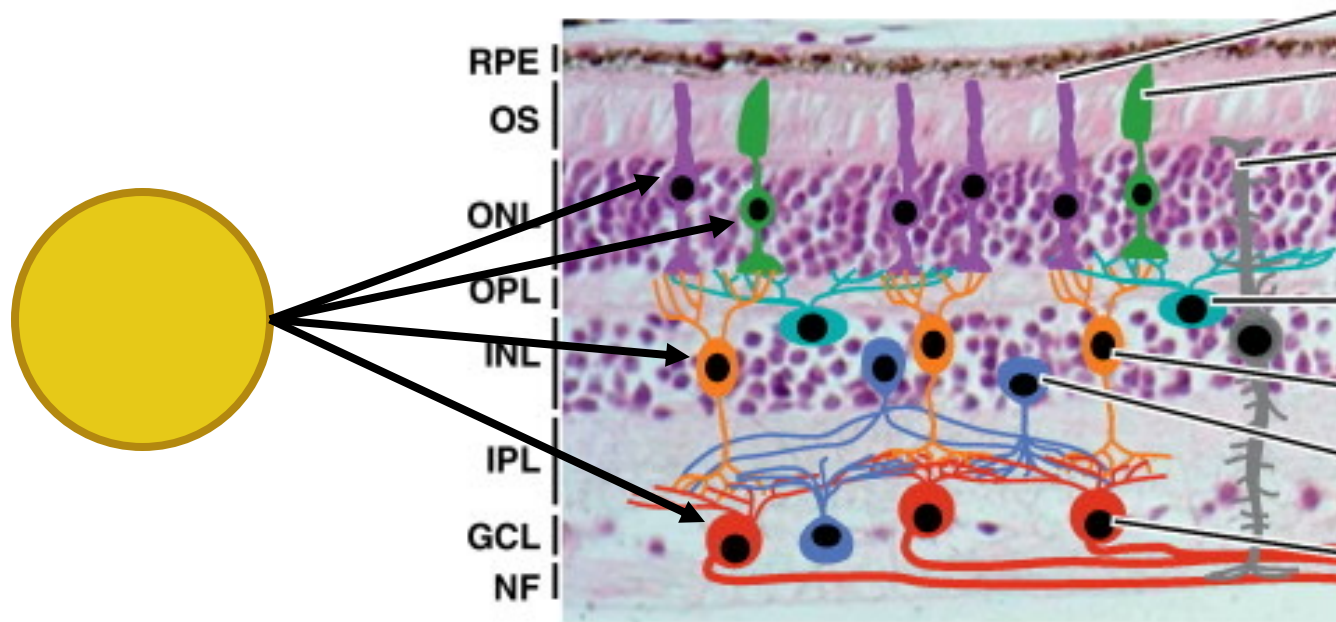


Cones
Bastonetes
Bipolares
Amácrinas
Horizontais
Ganglionares
Müller

Sequência de diferenciação Celular (gradiente)



Sequência de diferenciação Celular (gradiente)



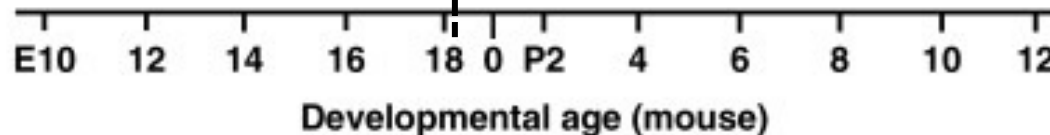
Amácrinas

Precoces

Cones
Horizontais
Ganglionares

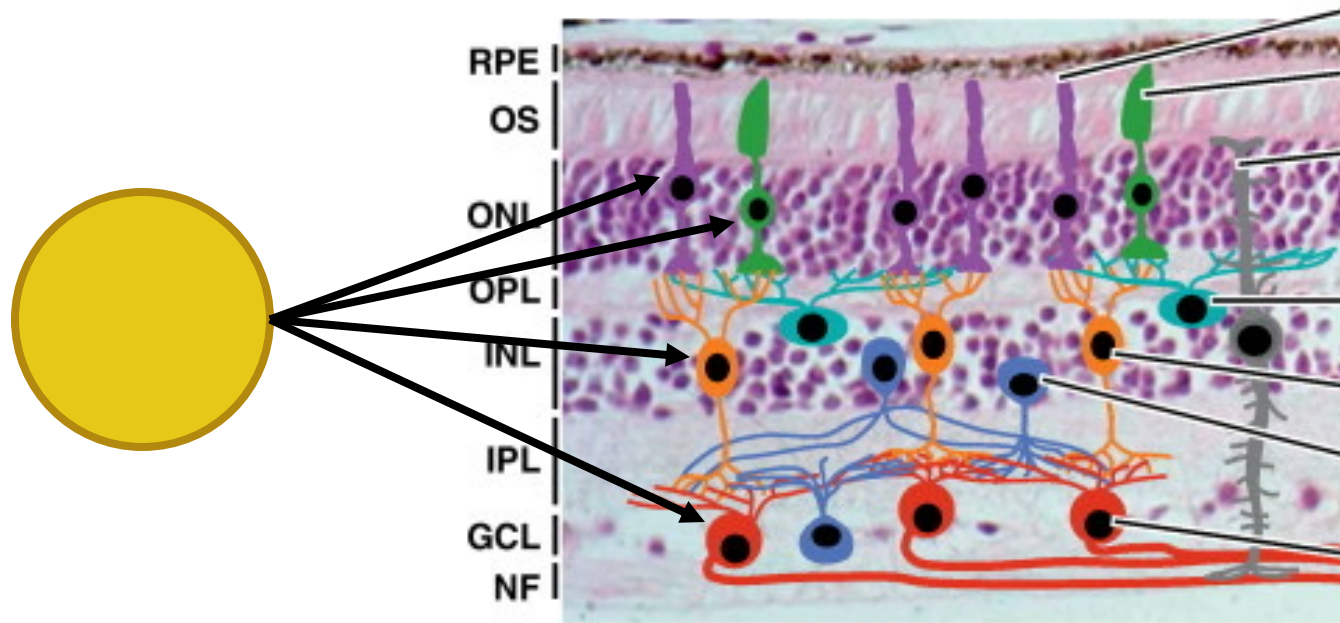
Bastonetes
Bipolares
Müller

Tardios



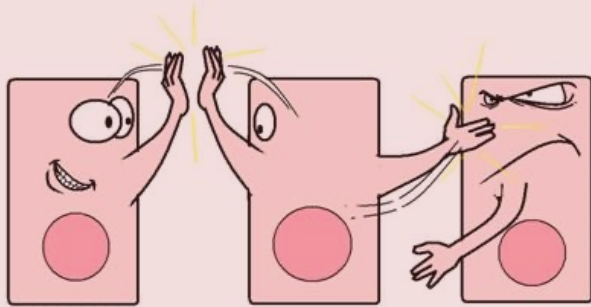
Determinação Celular dos Progenitores retinianos

Como acontece a escolha do destino celular?

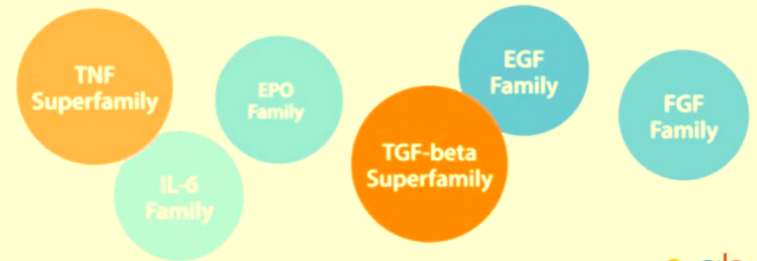


Determinação Celular dos Progenitores retinianos

Como acontece a escolha do perfil celular?



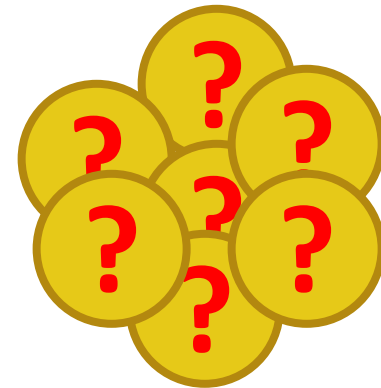
Interação Celular



Fatores de Crescimento



Fatores Secretados



Influência dos Fatores Secretados

PN0

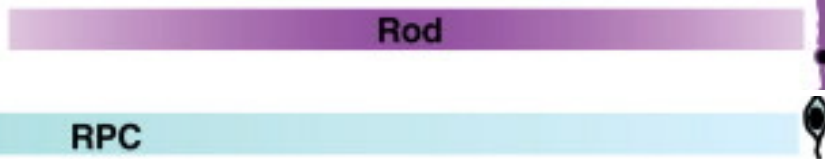
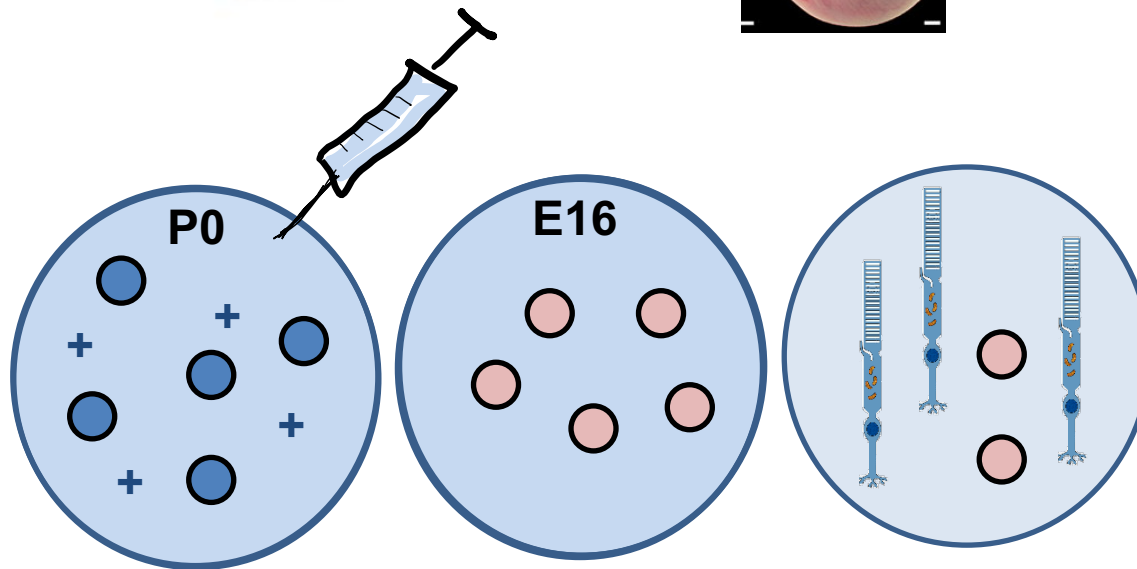
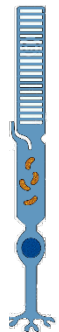


E16



Bastonetes
Bipolares
Amácrinas
Müller

Bastonetes
Bipolares
Amácrinas
Müller



E10 12 14 16 18 0 P2 4 6 8 10 12

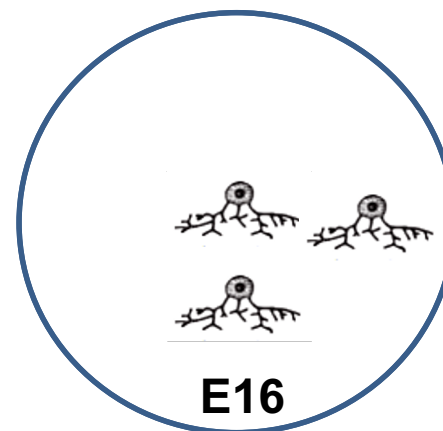
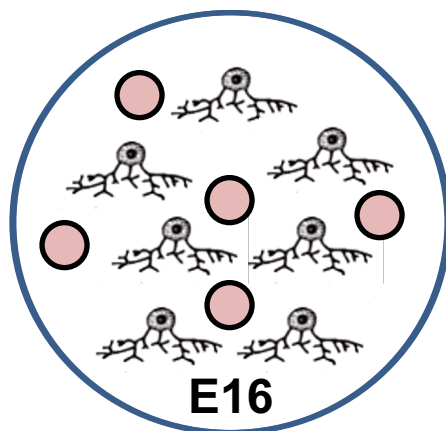
Developmental age (mouse)

Influência dos Fatores Secretados



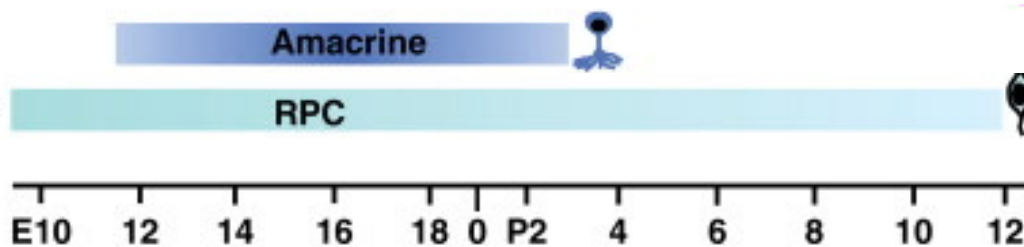
E16

Bastonetes
Bipolares
Amácrinas
Müller



Amacrine

RPC



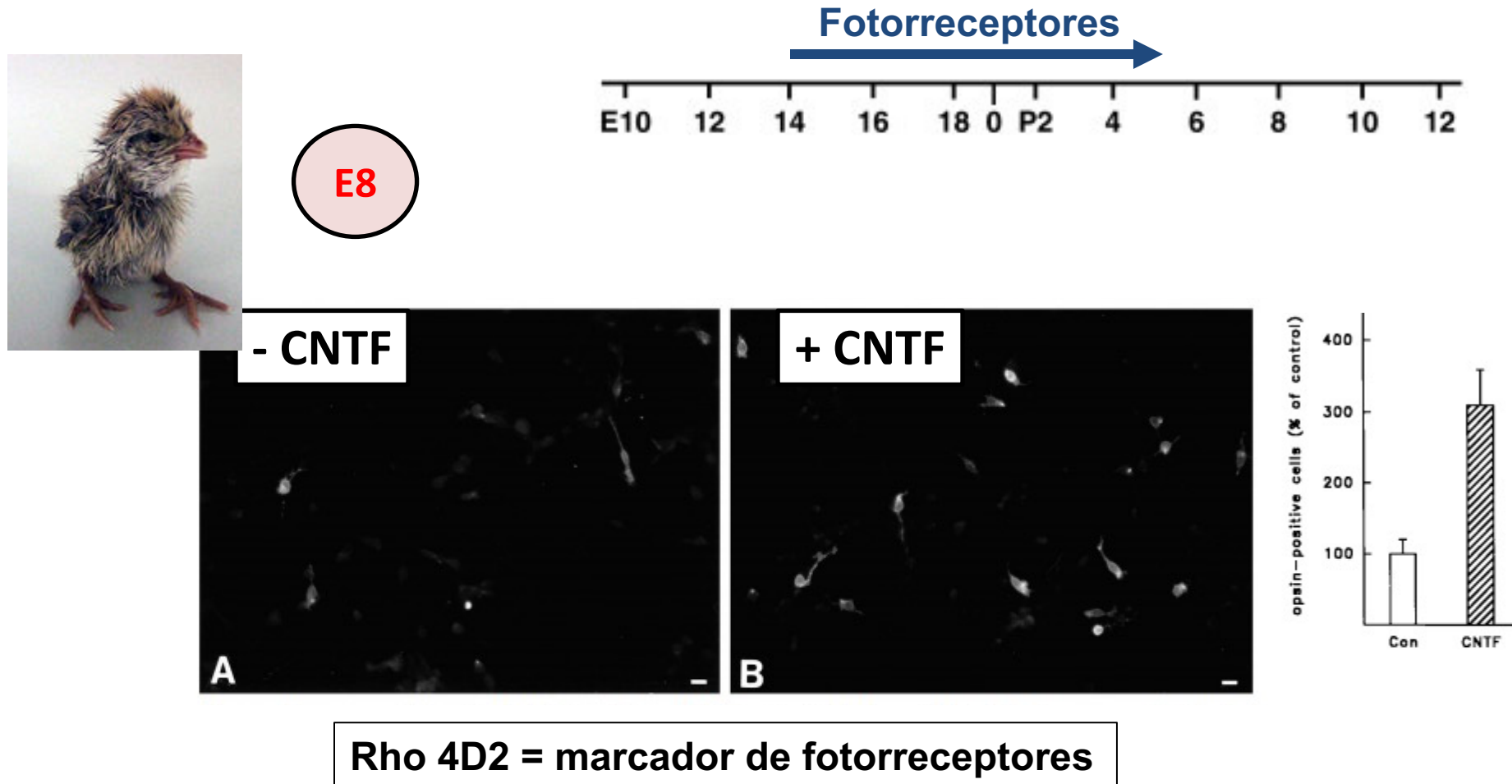
Developmental age (mouse)

**Células diferenciadas estimulam
feedback inibitório na produção
daquele mesmo tipo celular**

Belliveau MJ, Cepko CL. Development 1999, 126:555-566.

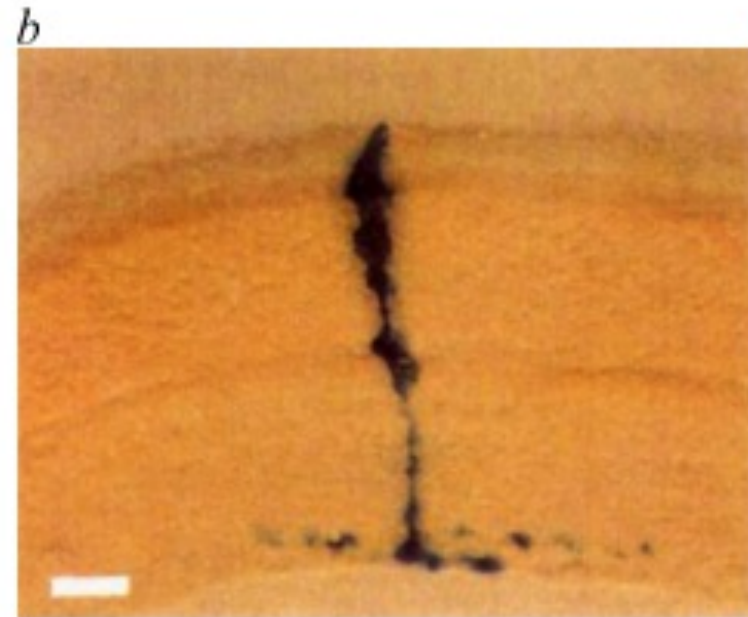
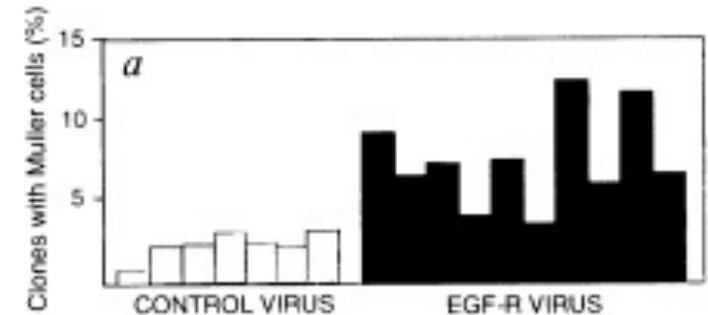
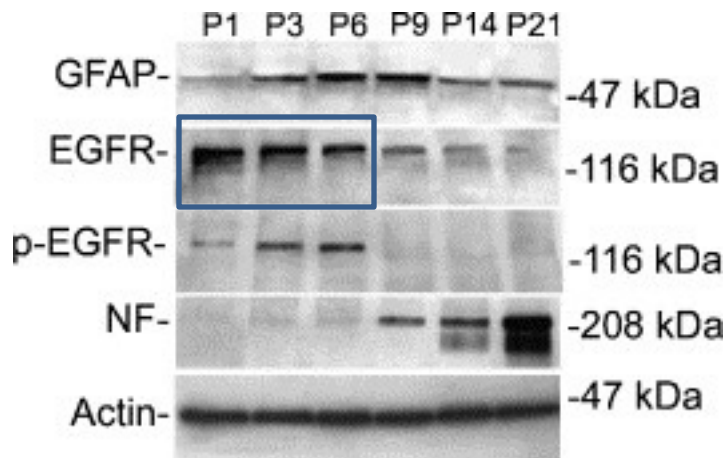
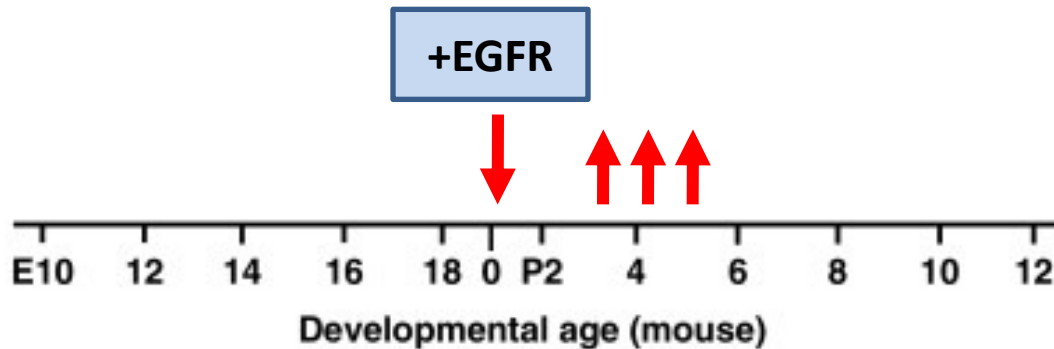
Influência dos Fatores de Crescimento

Fator neurotrófico ciliar (ciliary neurotrophic factor - **CNTF**) é secretado no início da fase de diferenciação dos fotorreceptores



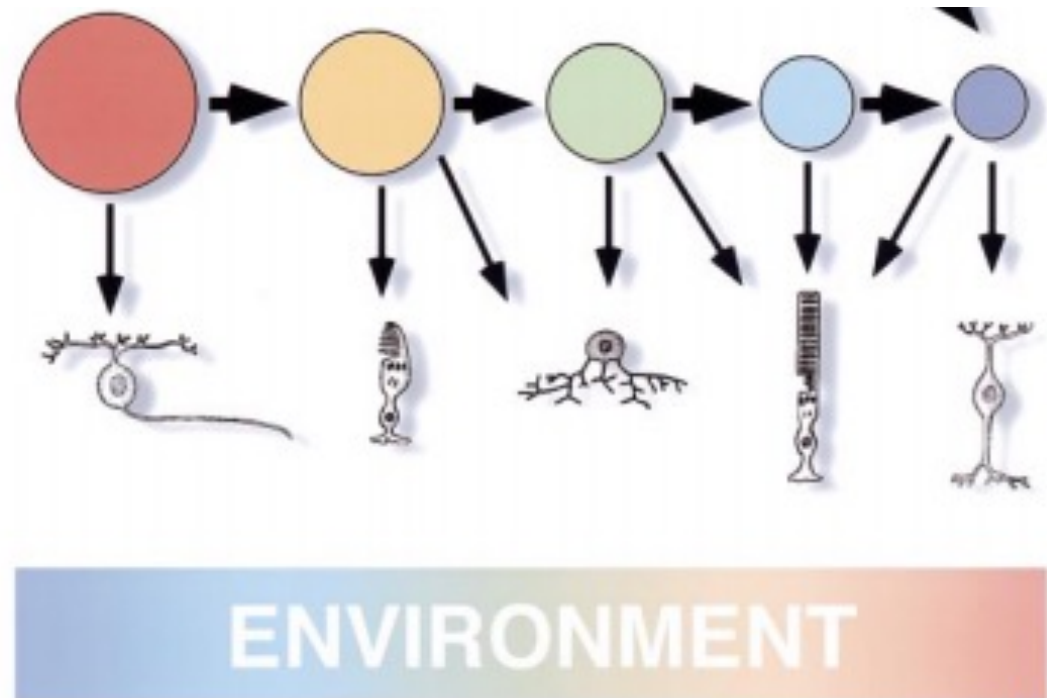
Influência dos Fatores de Crescimento

O Fator de Crescimento Epidermal (**EGF**) é conhecido por estimular a diferenciação das células gliais no período tardio de desenvolvimento



Os diferentes tipos celulares da retina se diferenciam em uma ordem cronológica conservada

Interação Celular
Fatores de Crescimento
Fatores Secretados
=
Tipos diferentes de neurônios e glia

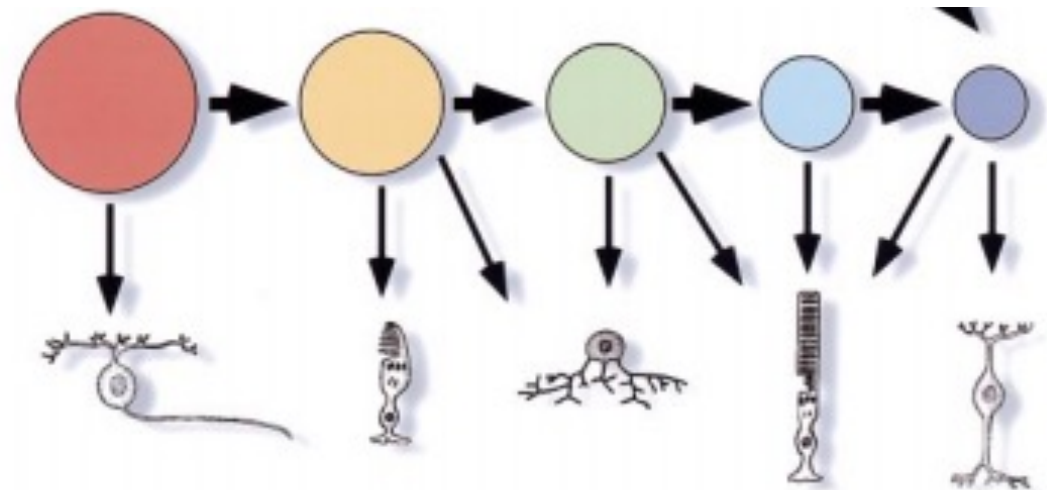


Aula: Desenvolvimento e Regeneração da Retina

Parte 3 - Desenvolvimento da Retina

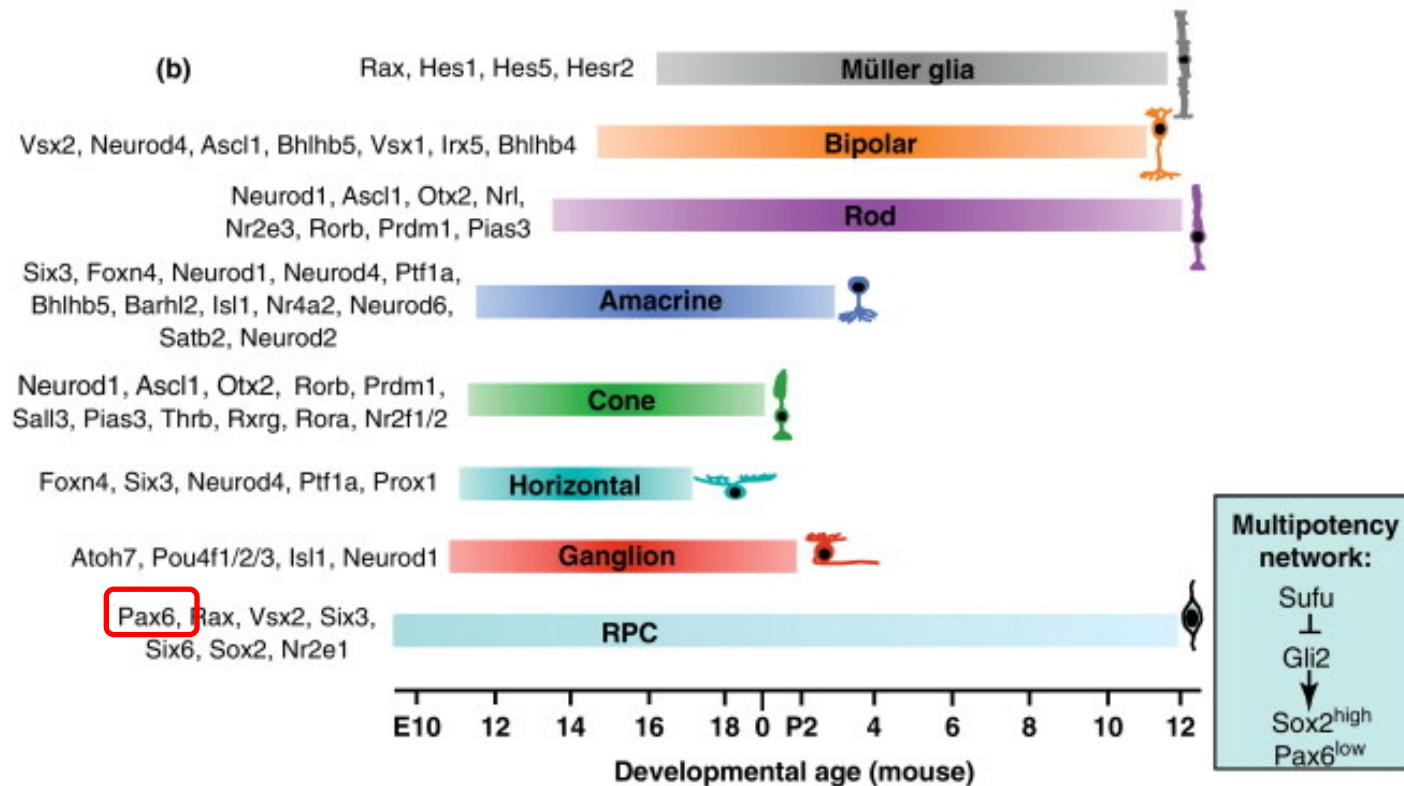
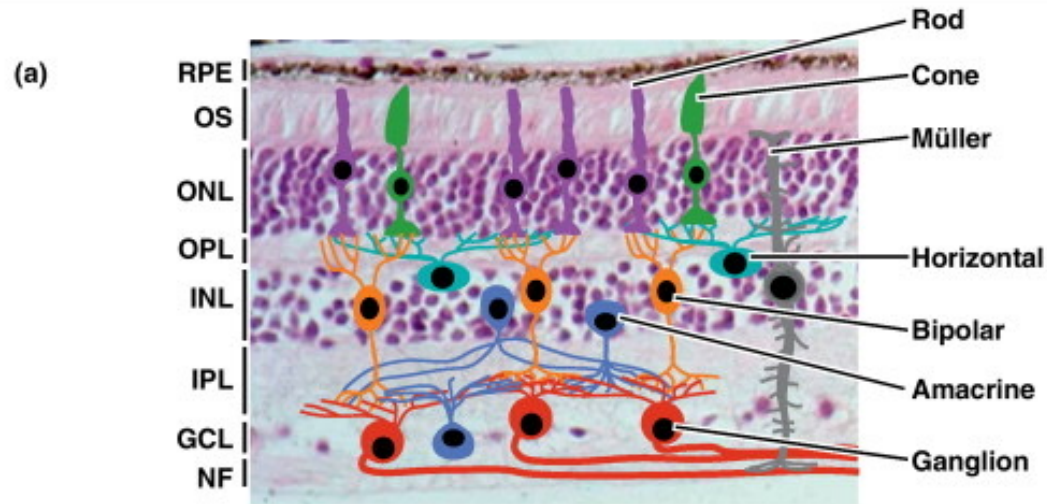
Os diferentes tipos celulares da retina se diferenciam em uma ordem cronológica conservada

Interação Celular
Fatores de Crescimento
Fatores Secretados



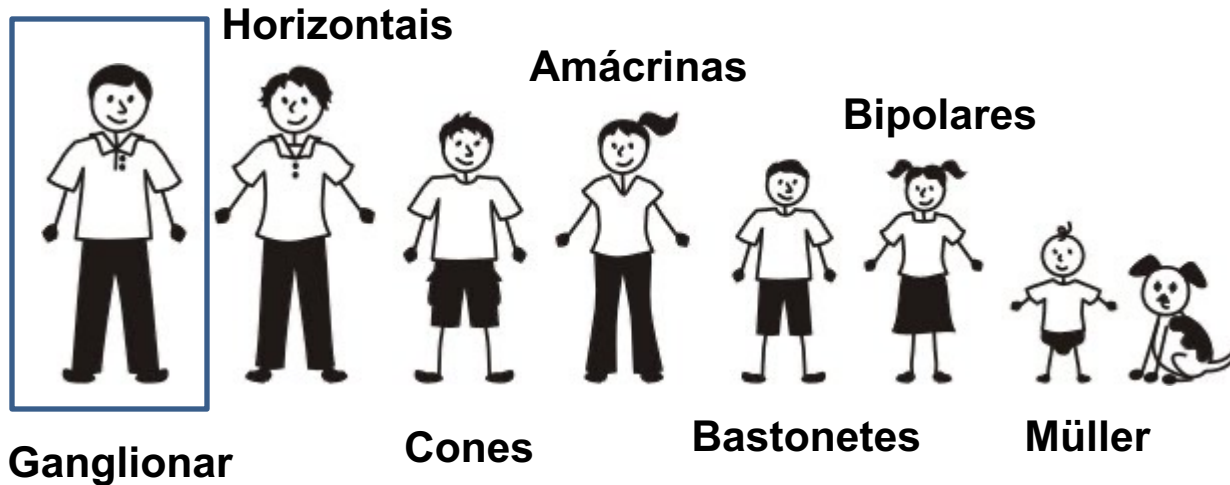
ENVIRONMENT

Current Opinion in Neurobiology



Diferenciação das células retinianas

Primogênito: Células Ganglionares



Atoh7, Pou4f1/2/3, Isl1, Neurod1

Ganglion



Pax6, Rax, Vsx2, Six3,
Six6, Sox2, Nr2e1

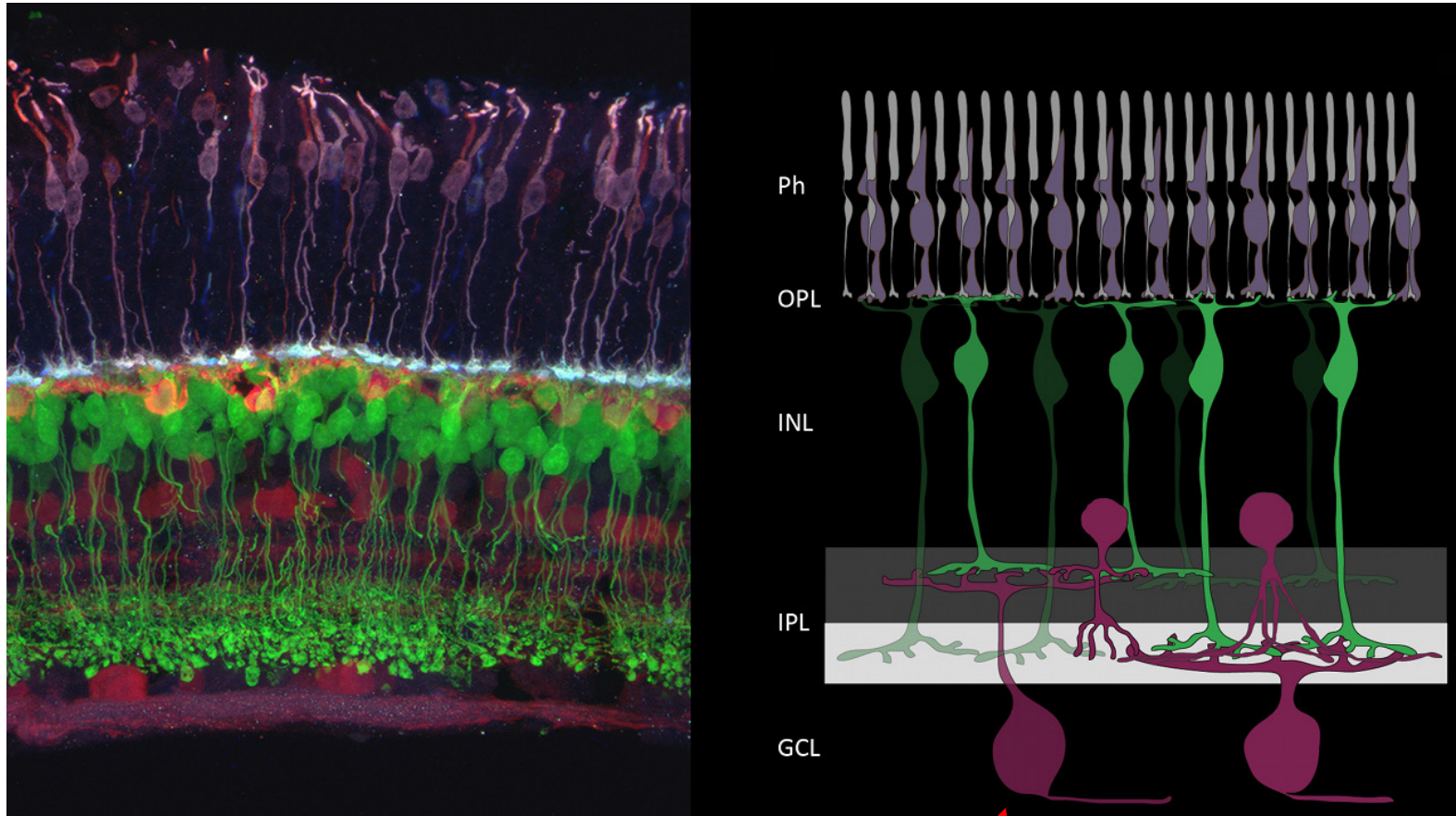
RPC

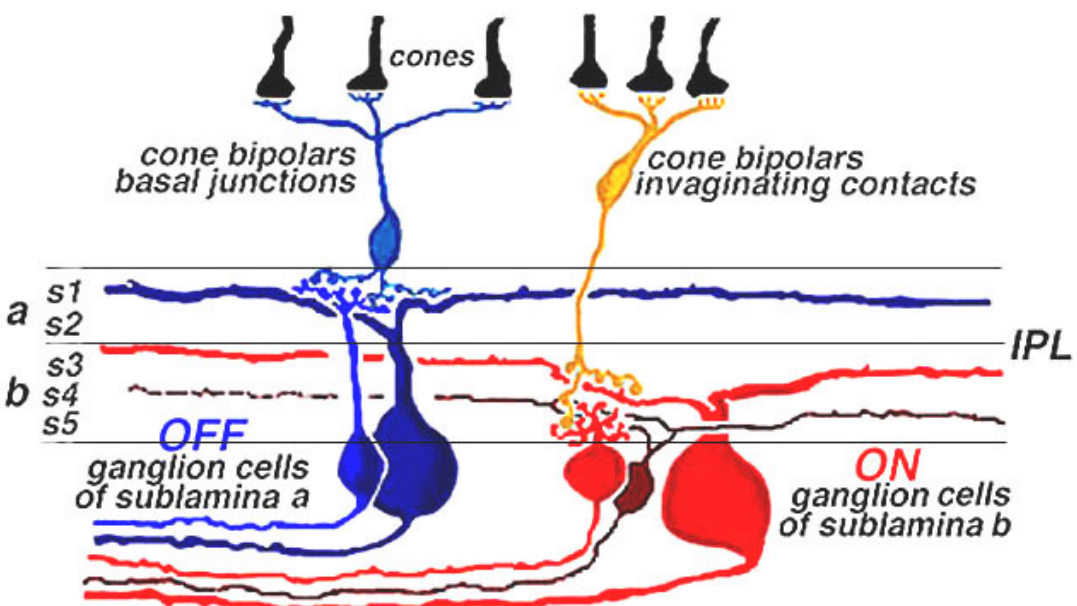


E10 12 14 16 18 0 P2 4 6 8 10 12

Developmental age (mouse)

As células ganglionares são responsáveis por coletar todas as informações interpretadas pelos neurônios retinianos e encaminhar para os centros visuais

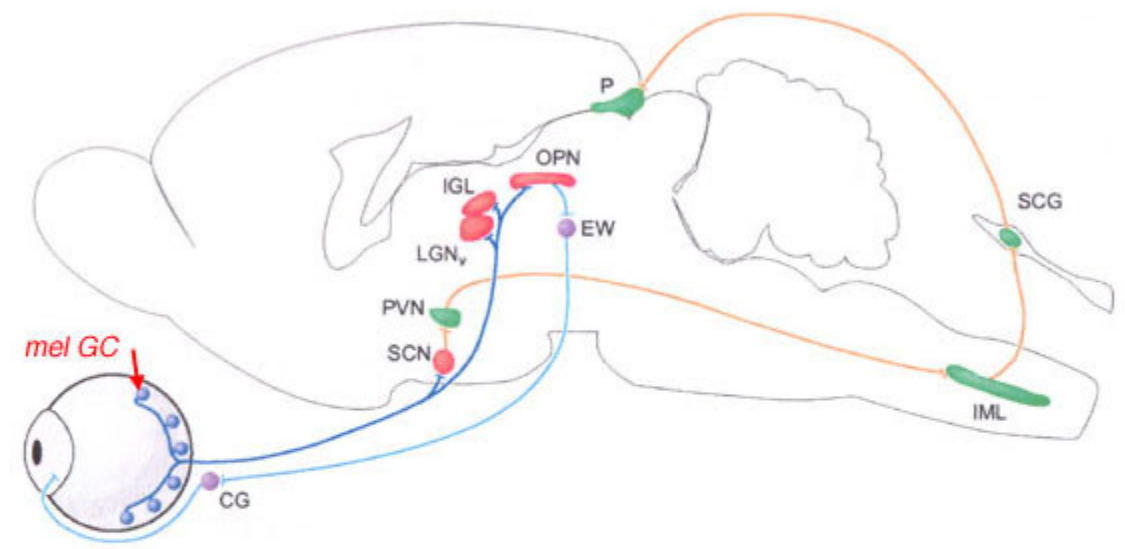
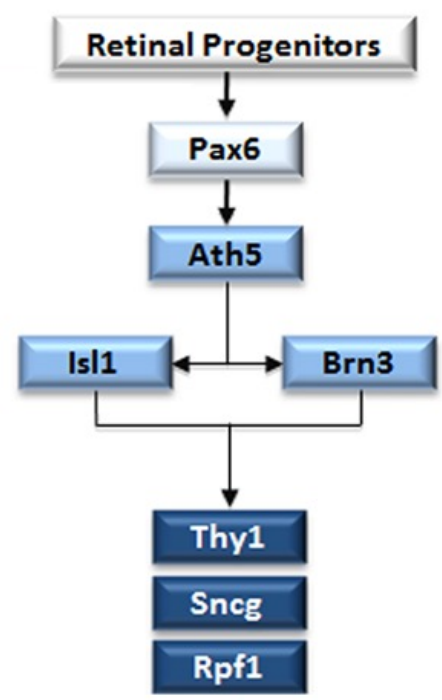




Nelson, Famiglietti & Kolb, 1978

Interpretam:

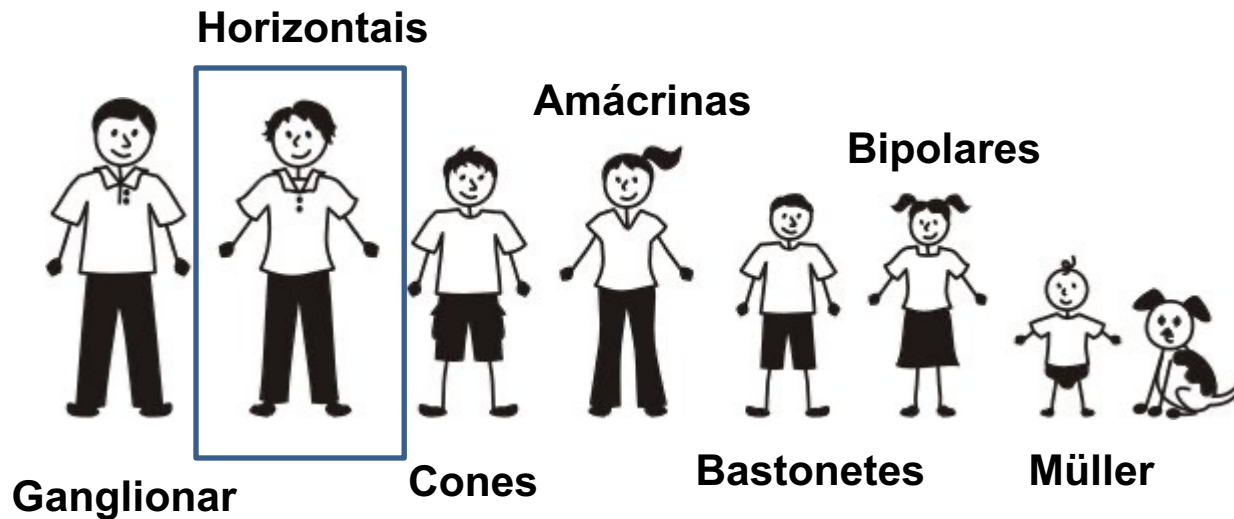
- Alterações de brilho
- Alterações de formas
- Movimentação



Núcleo geniculado lateral e colículo superior

Diferenciação das células retinianas

Células Horizontais



Foxn4, Six3, Neurod4, Ptf1a, Prox1

Horizontal



Atoh7, Pou4f1/2/3, Isl1, Neurod1

Ganglion



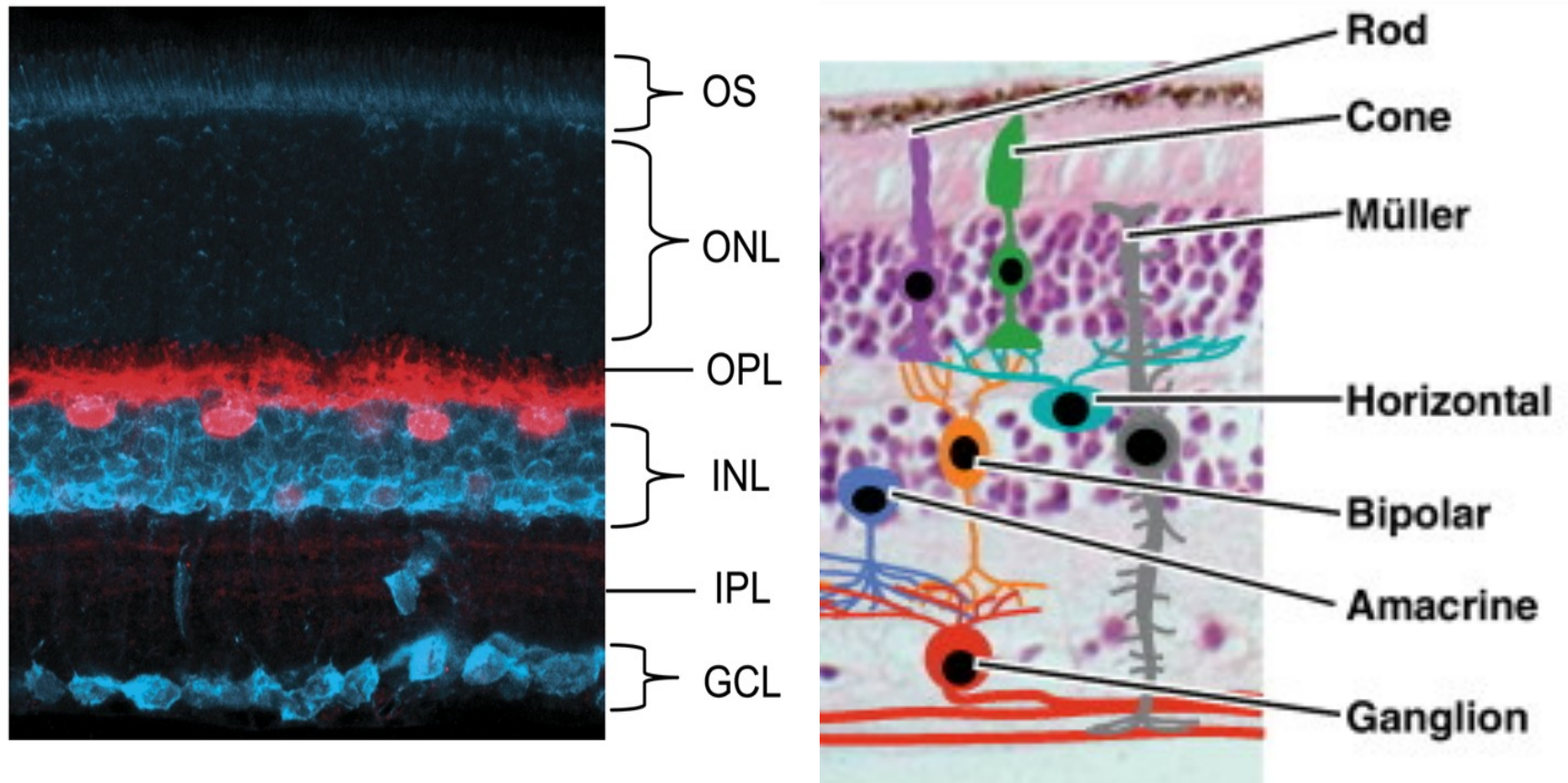
Pax6, Rax, Vsx2, Six3,
Six6, Sox2, Nr2e1

RPC



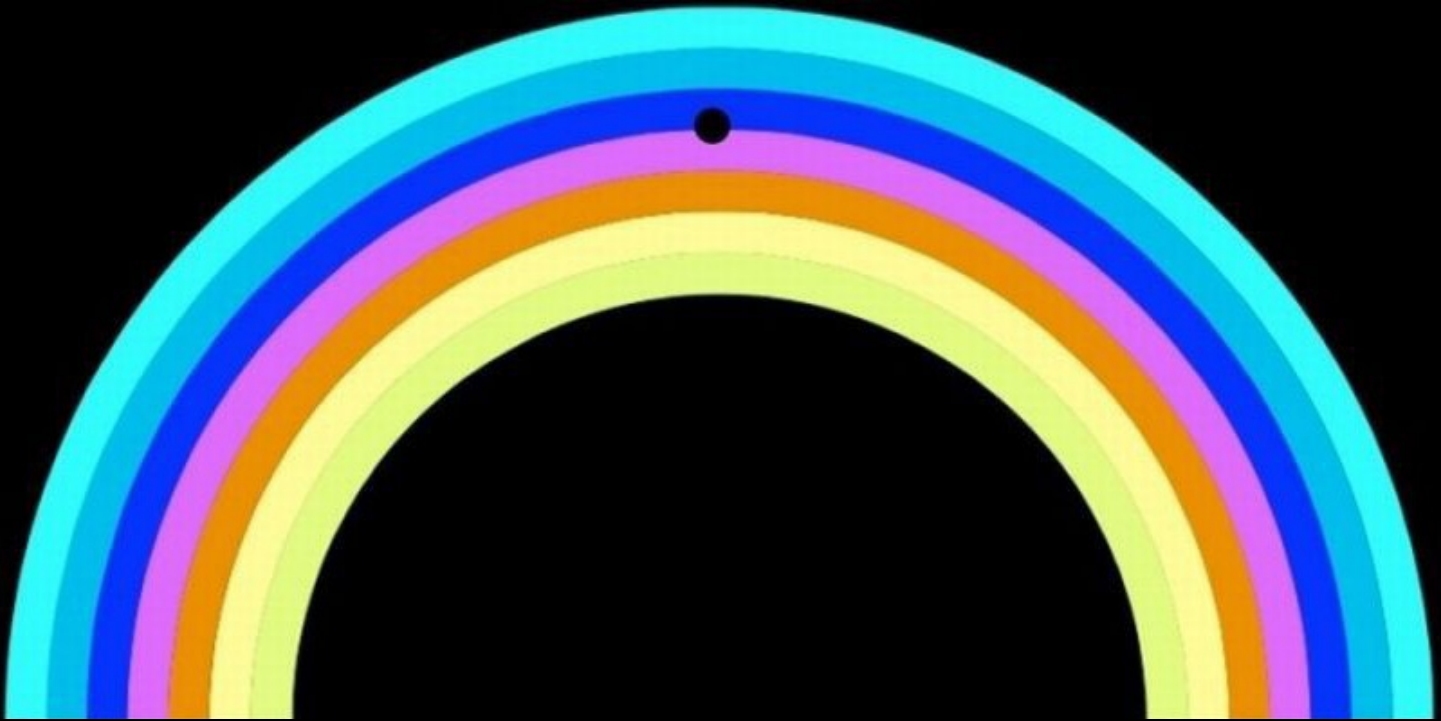
E10 12 14 16 18 0 P2 4 6 8 10 12

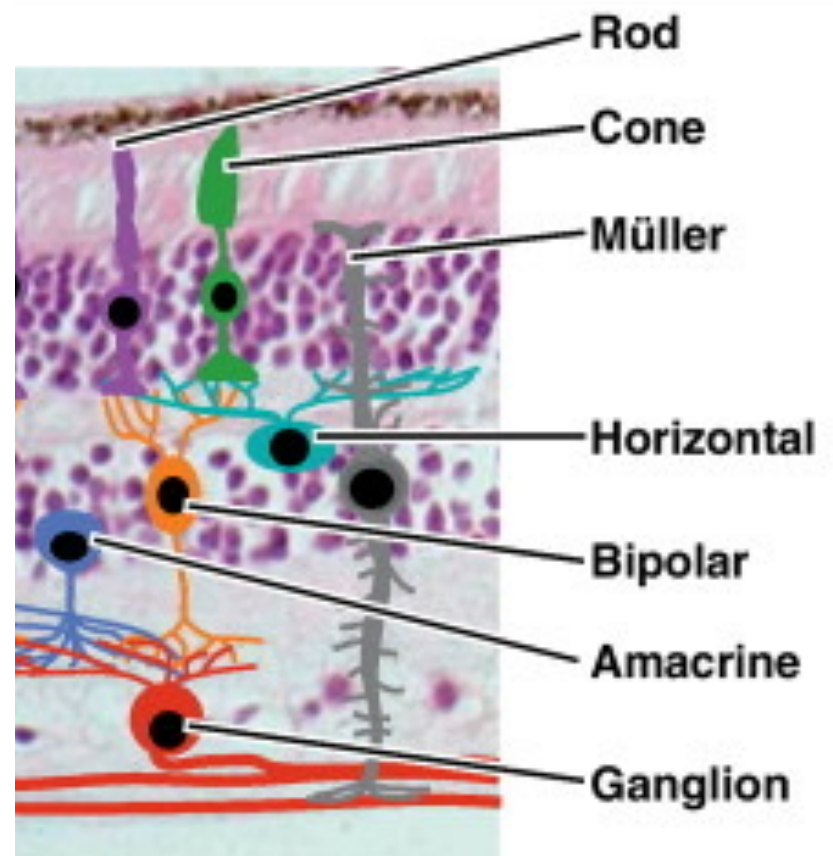
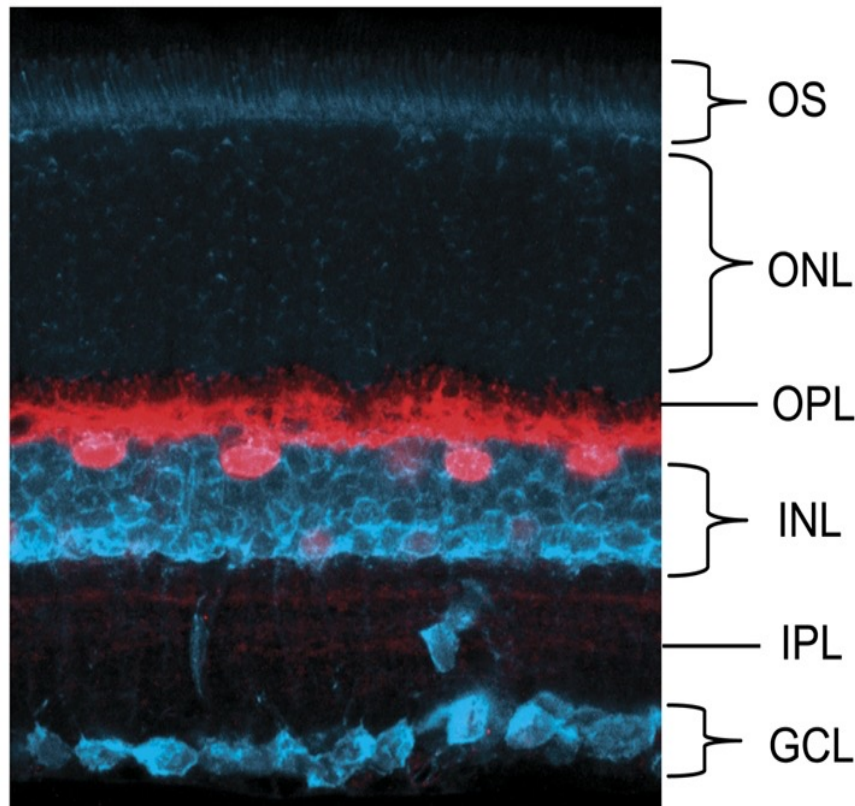
Developmental age (mouse)



Modulam os sinais gerados pelos fotorreceptores:

- Promovem a interação dos fotorreceptores
- Refinam as informações de cores opostas (ilusões)

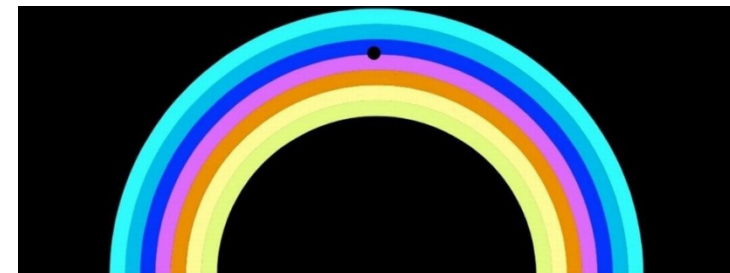


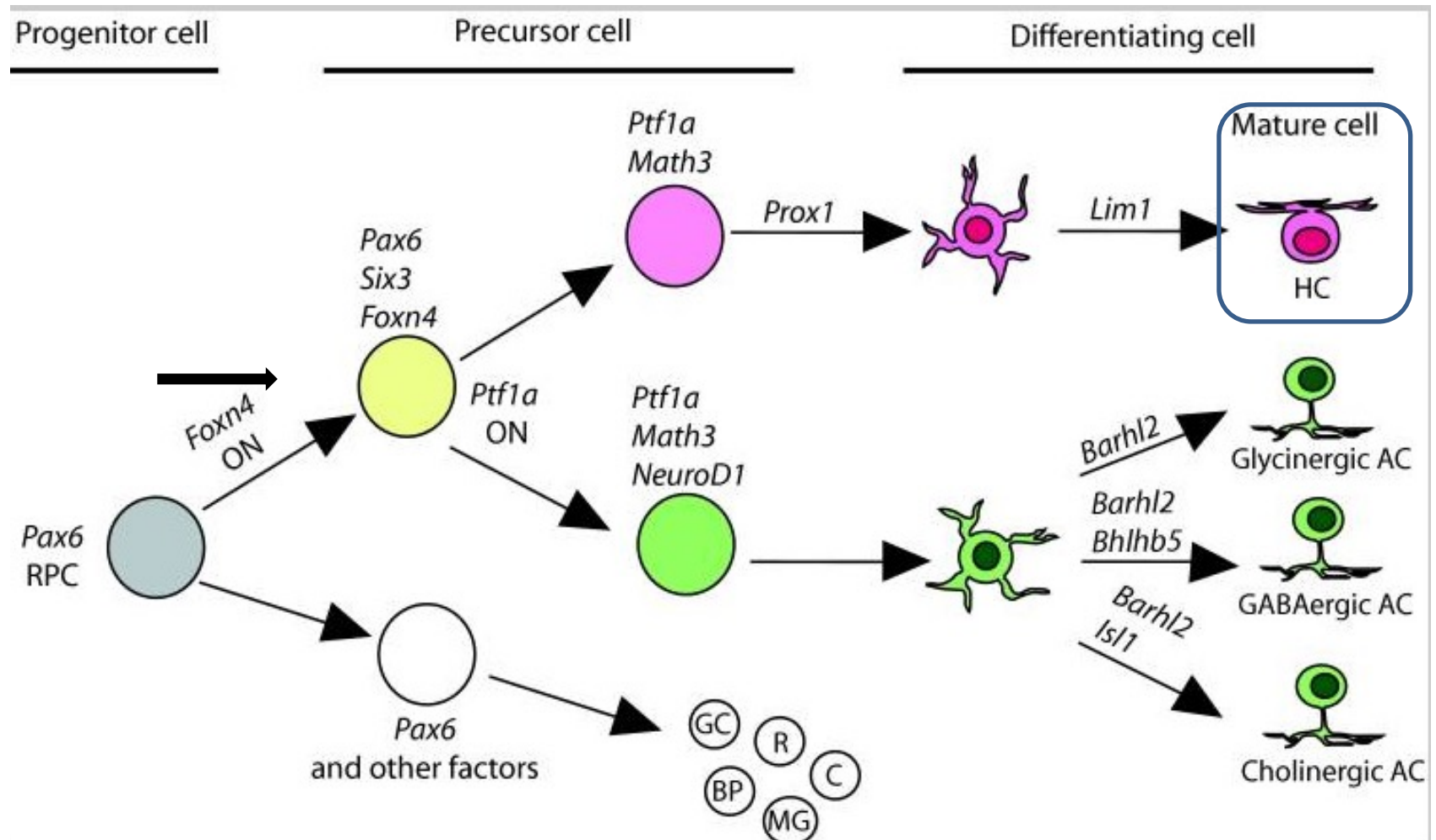


Modulam os sinais gerados pelos fotorreceptores:

- Promovem a **interação dos fotorreceptores**
- Refinam as informações de cores opostas (ilusões)

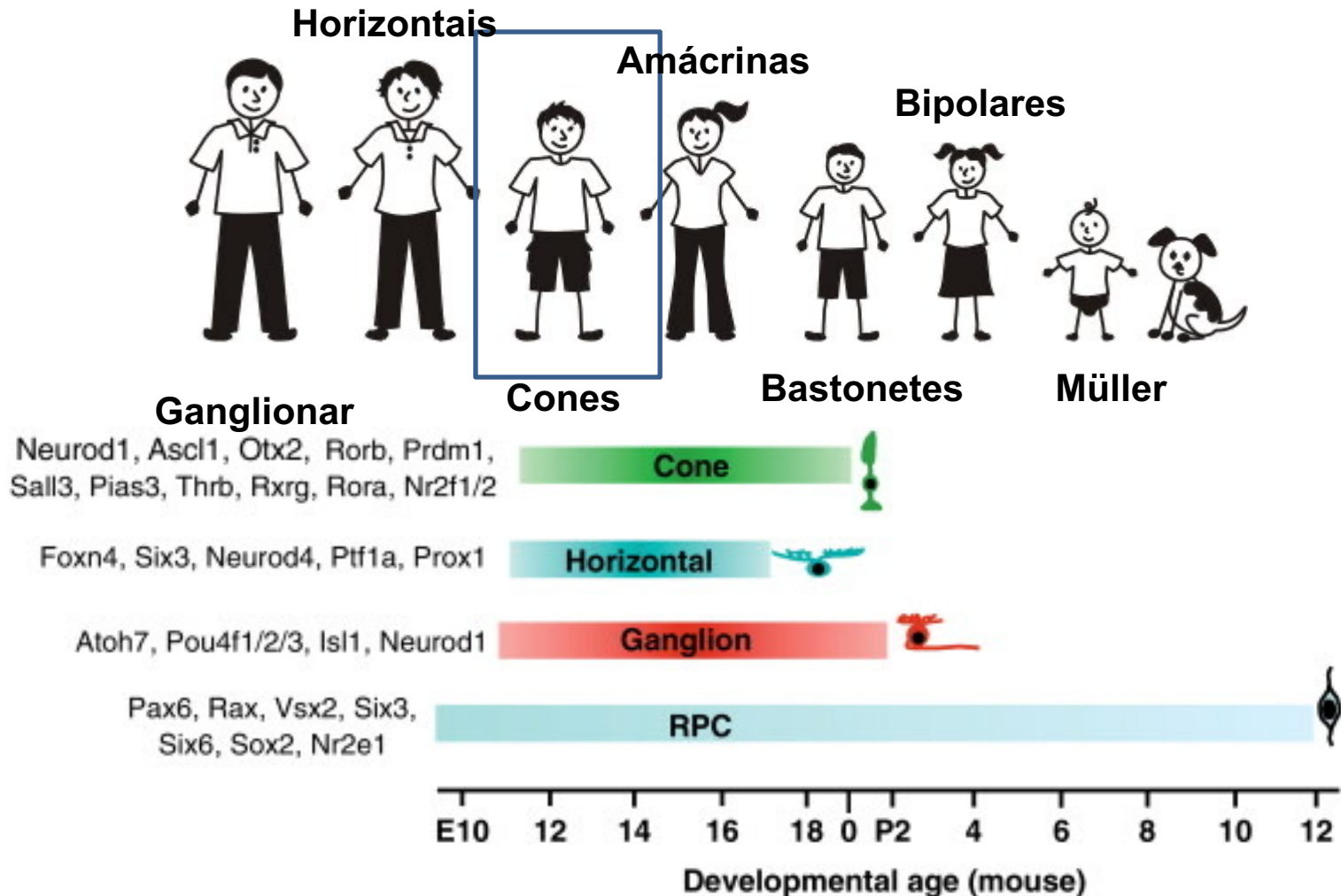
*Poché & Reese. Development. 2009 Jul 1;
136(13): 2141–2151.*





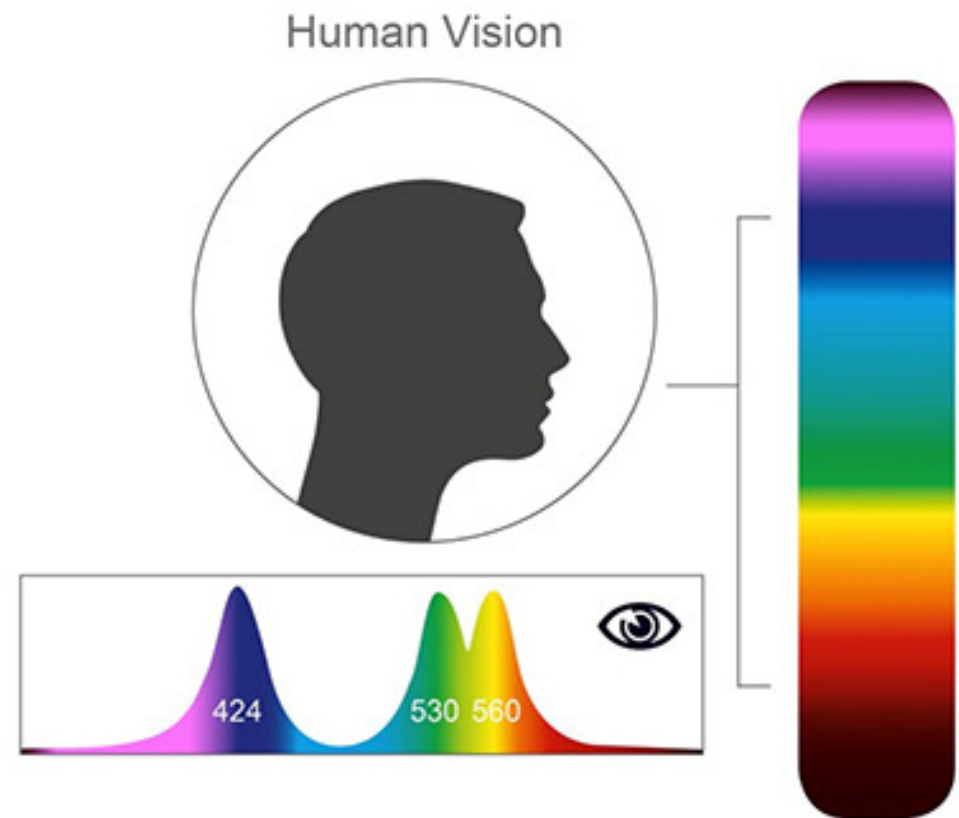
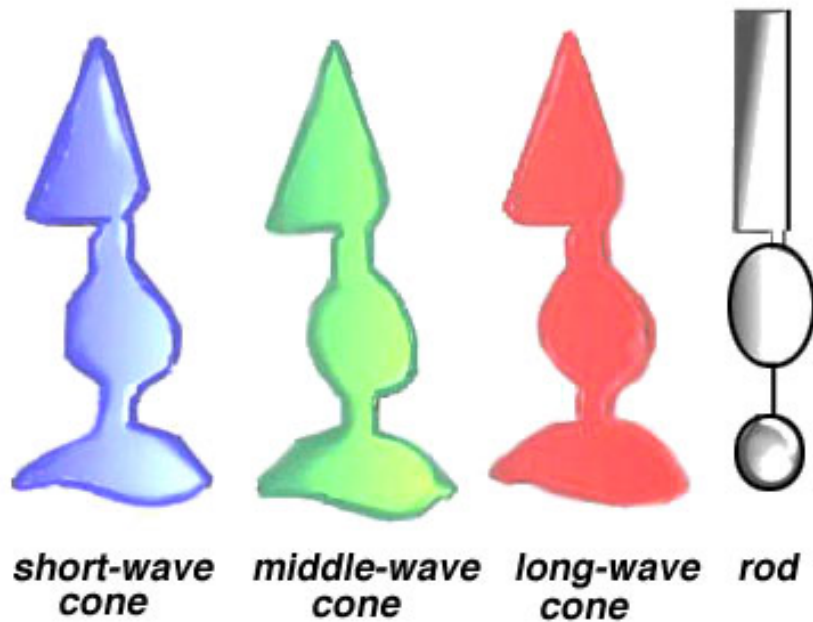
Diferenciação das células retinianas

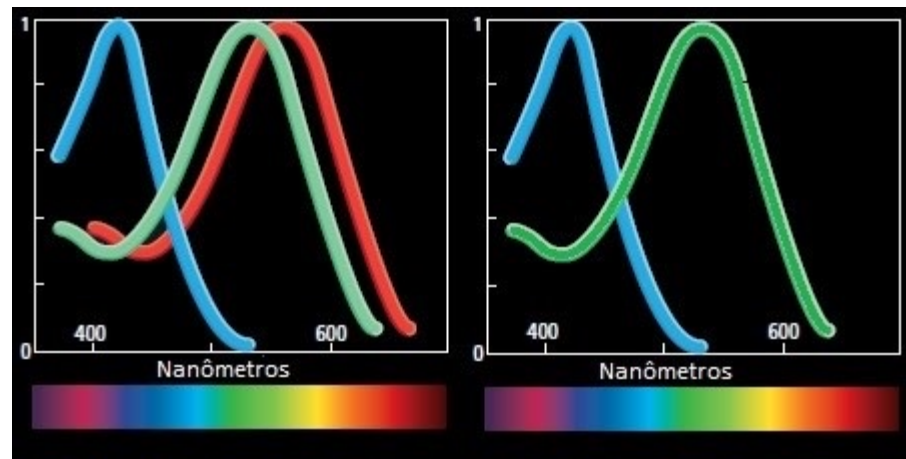
Células Fotorreceptoras: Cones

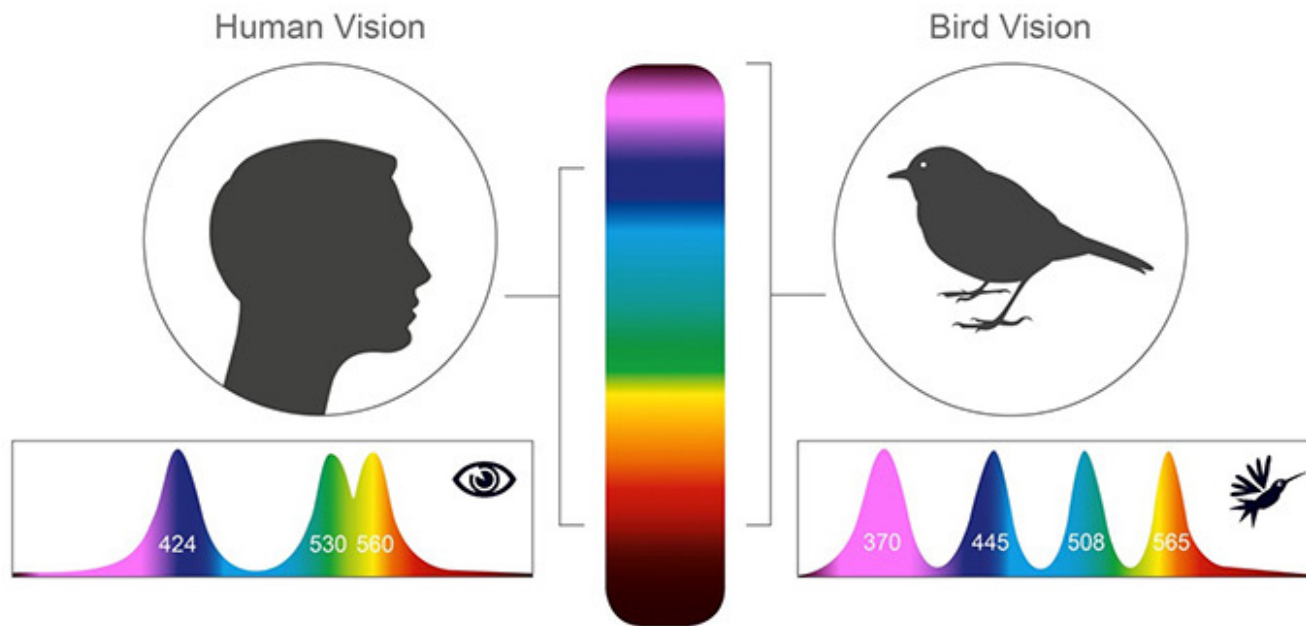


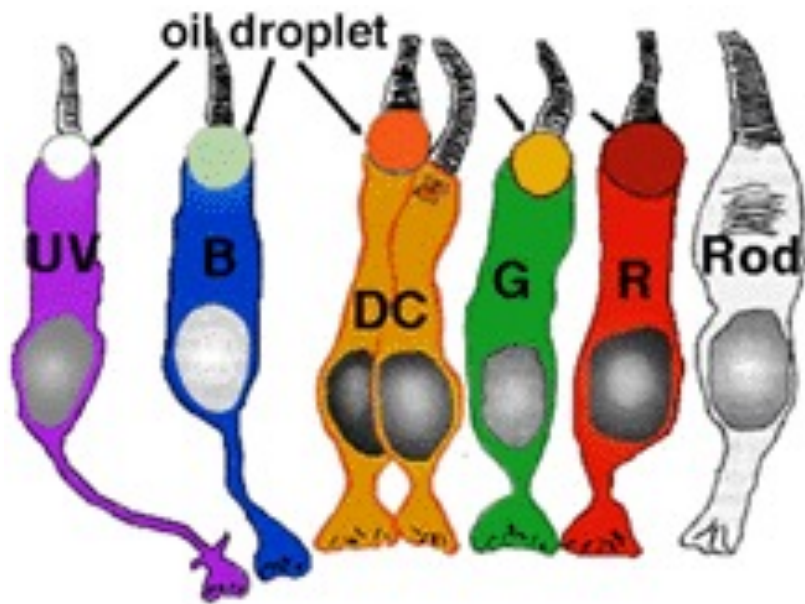
Células Fotorreceptoras: Cones

Visão de Cores









Daltonismo: Deficiência no pigmento dos cones (protanopia – pigmento vermelho)

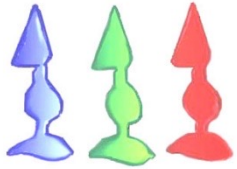
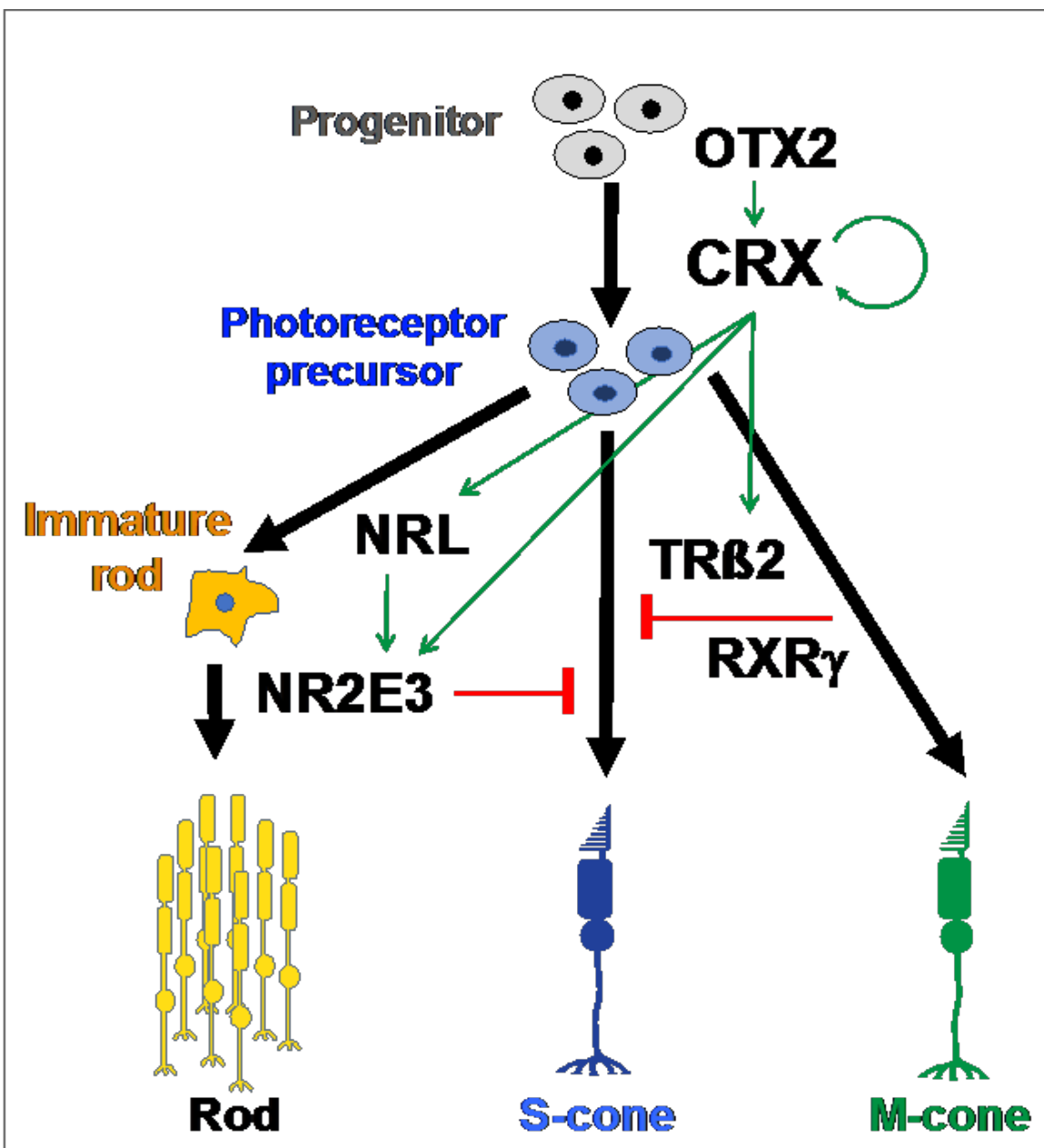
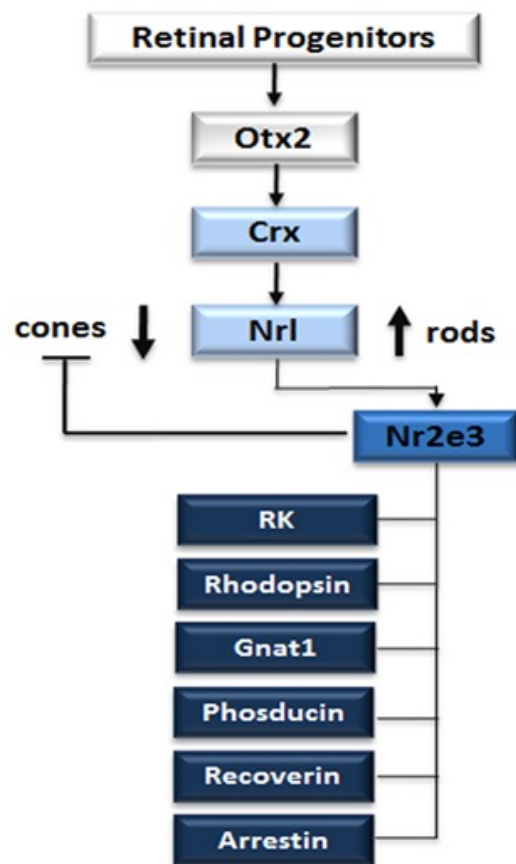




Imagem Negativa: os fotorreceptores se tornam adaptados e param de sinalizar, assim os fotorreceptores laterais sinalizam fortemente quando troca o fundo

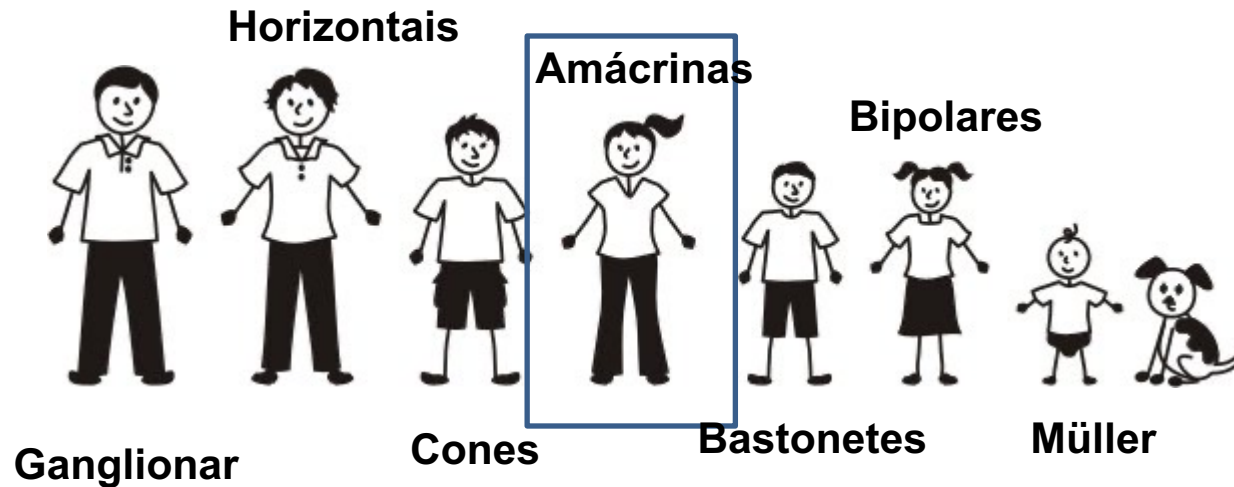


Imagem Negativa: os fotorreceptores se tornam adaptados e param de sinalizar, assim os fotorreceptores laterais sinalizam fortemente quando troca o fundo



Diferenciação das células retinianas

Células Amácrinas



Six3, Foxn4, Neurod1, Neurod4, Ptf1a,
Bhlhb5, Barhl2, Isl1, Nr4a2, Neurod6,
Satb2, Neurod2

Amacrine



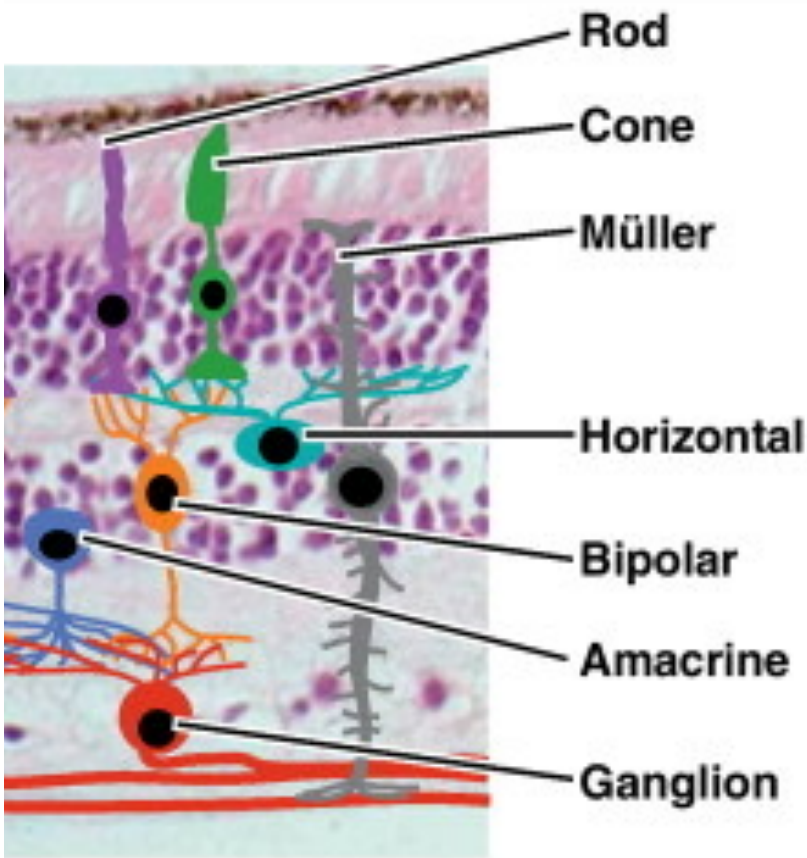
Pax6, Rax, Vsx2, Six3,
Six6, Sox2, Nr2e1

RPC

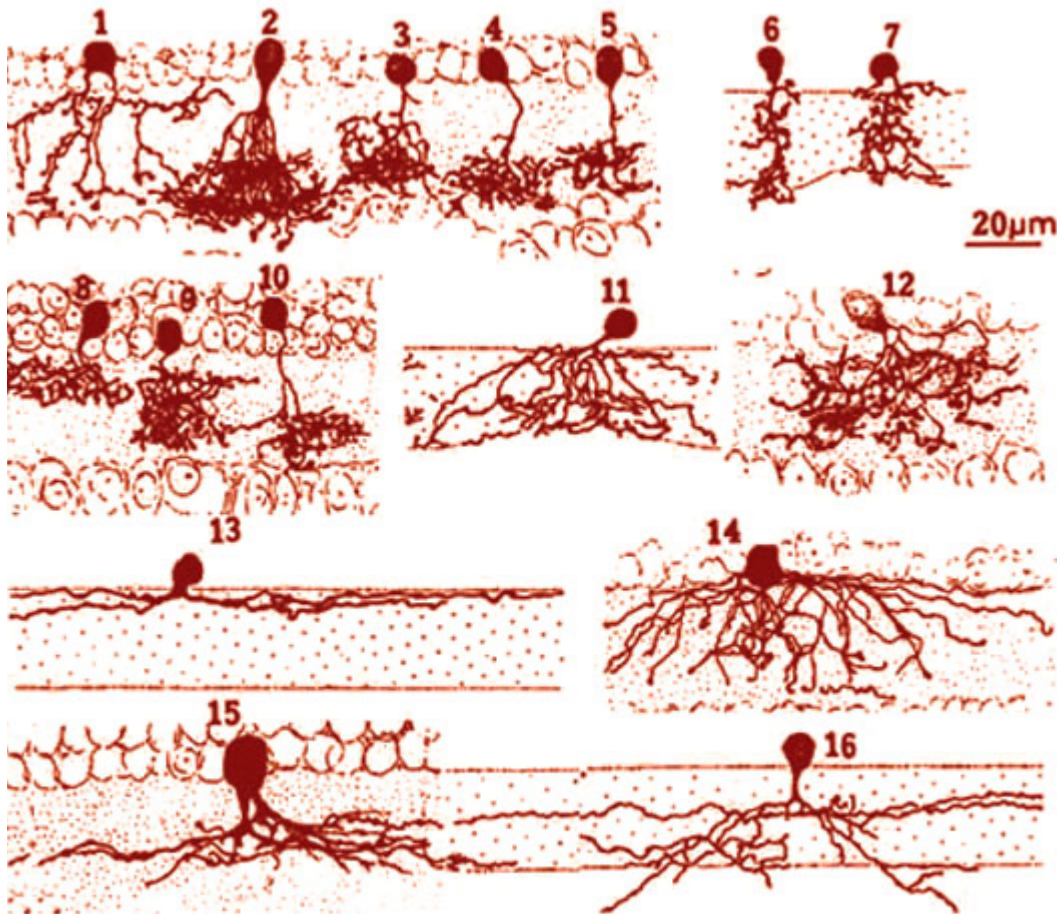


E10 12 14 16 18 0 P2 4 6 8 10 12

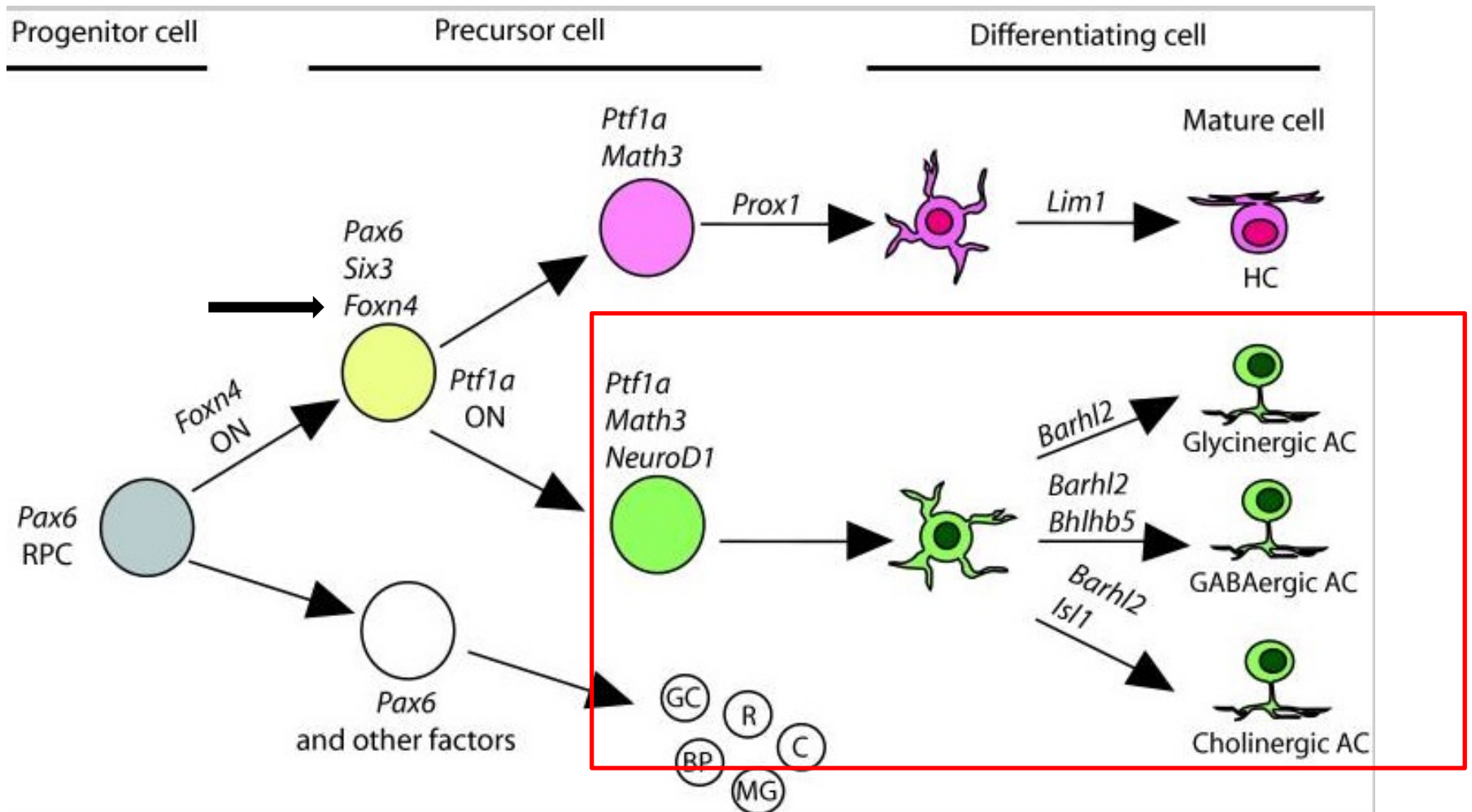
Developmental age (mouse)



Modulam as informações da
cadeia
Fotorreceptor – bipolar-
ganglionar

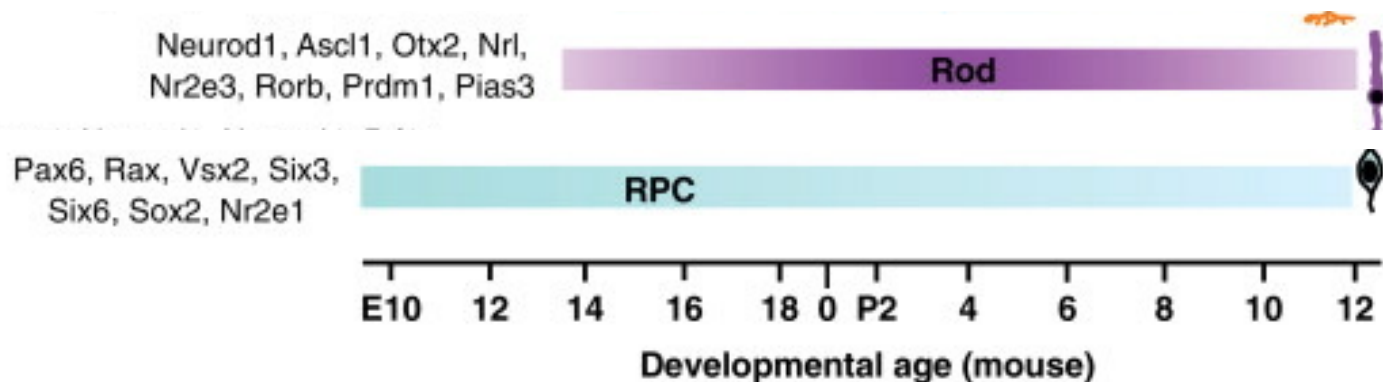
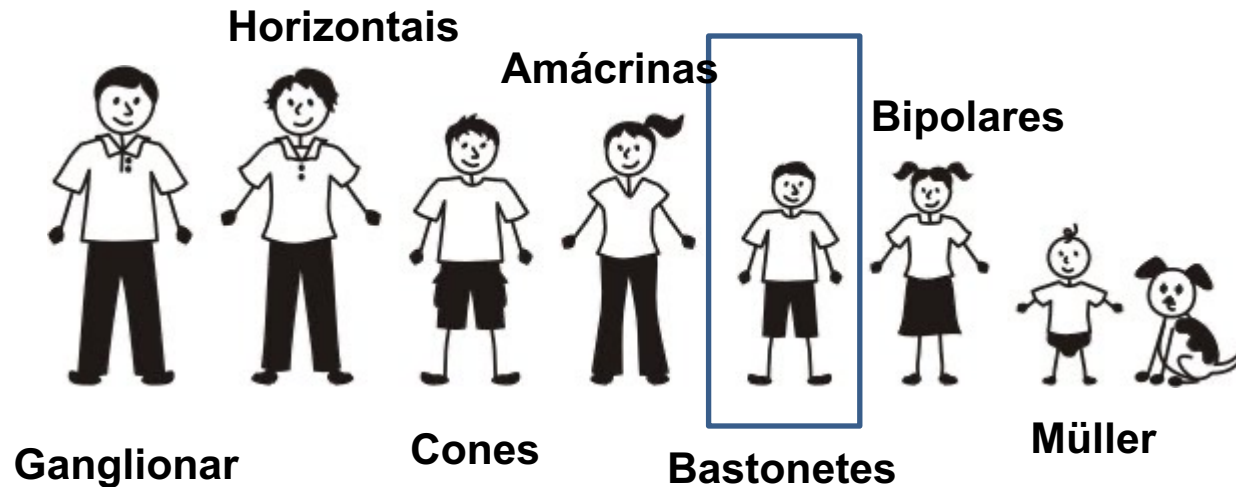


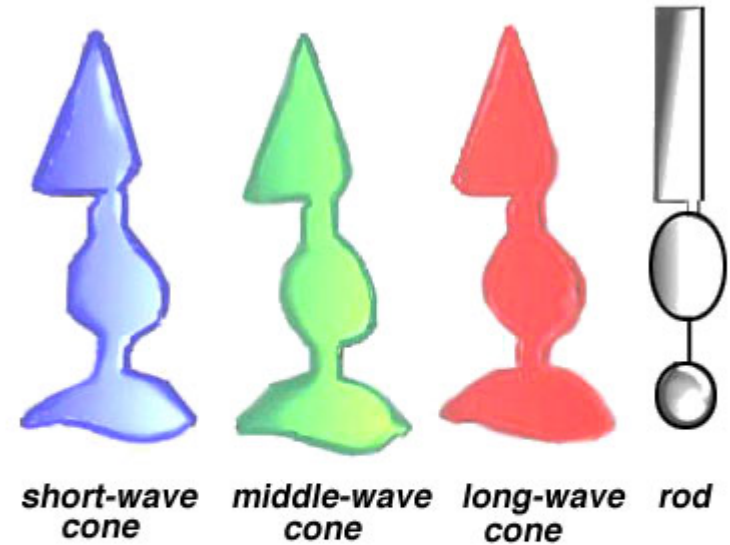
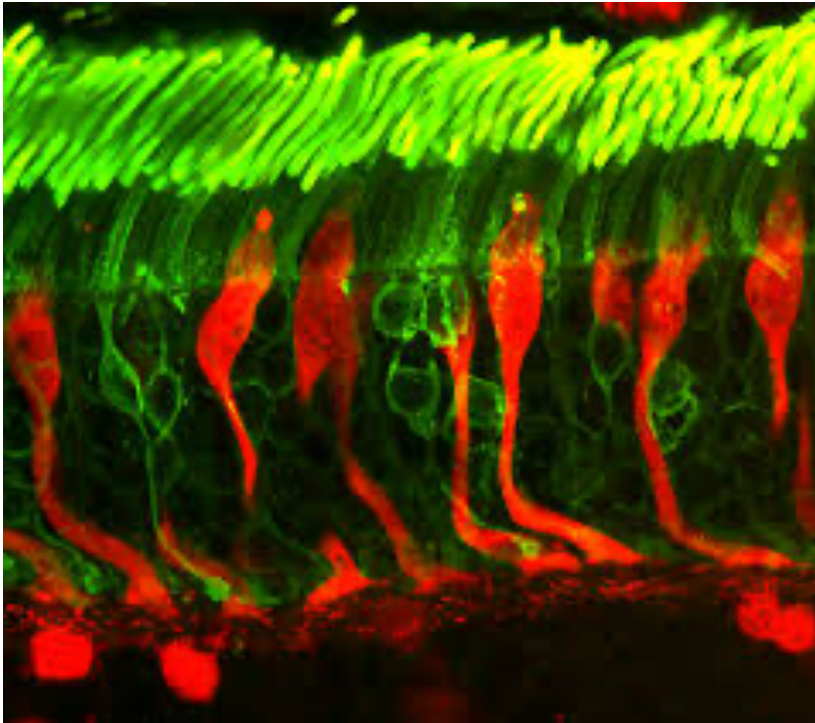
g. 6. Stratification patterns of small and medium-field amacrine cells in primate retina (from Polyak, 1941).



Diferenciação das células retinianas

Células Fotorreceptoras: Bastonetes

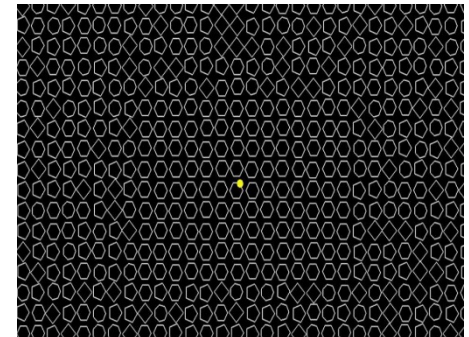


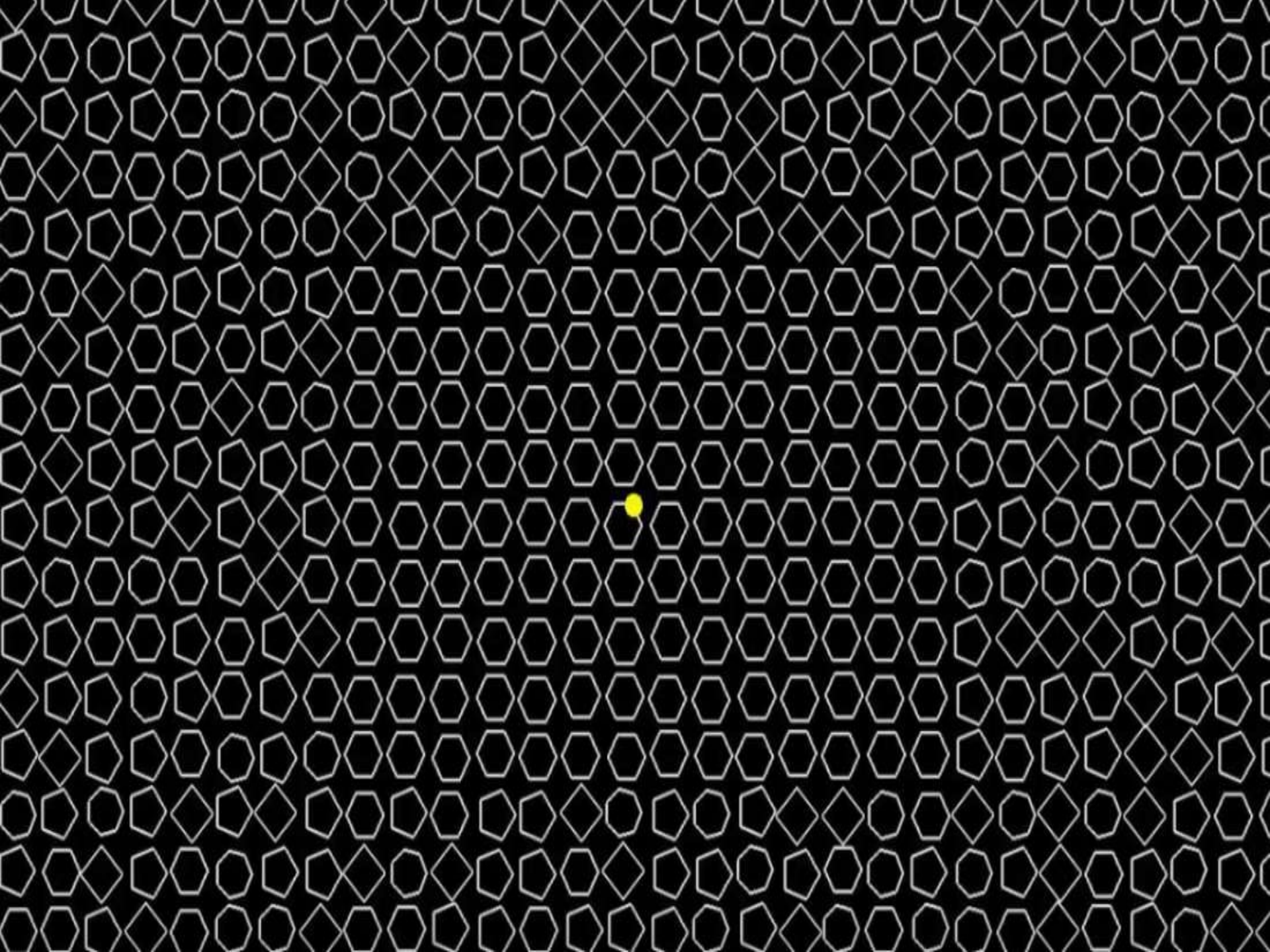


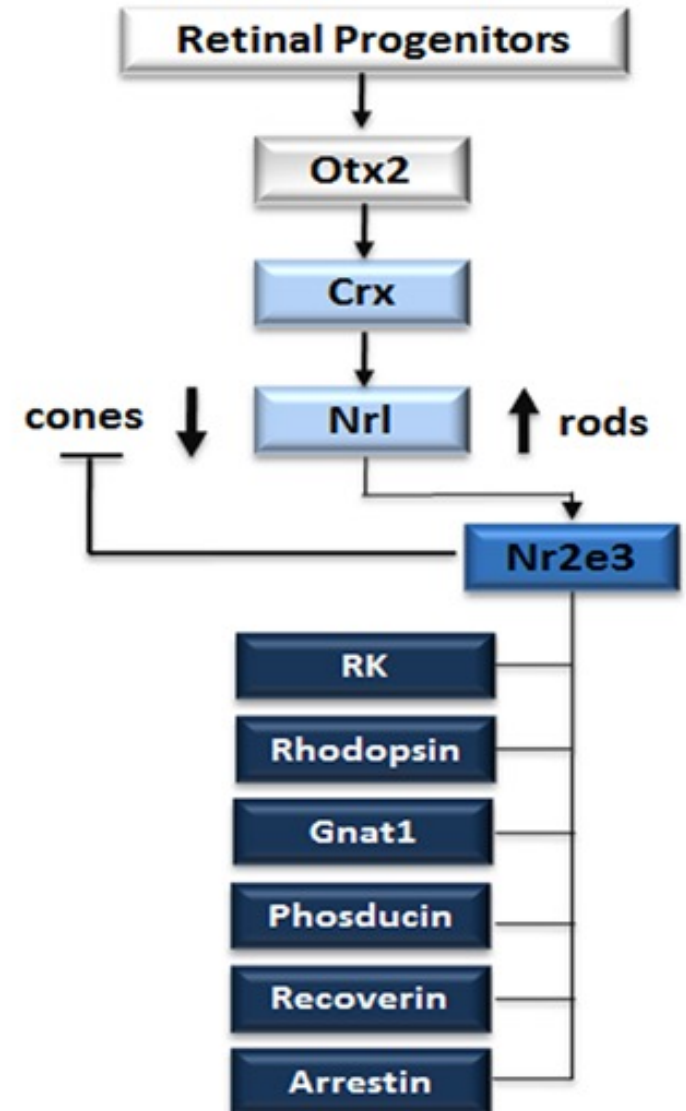
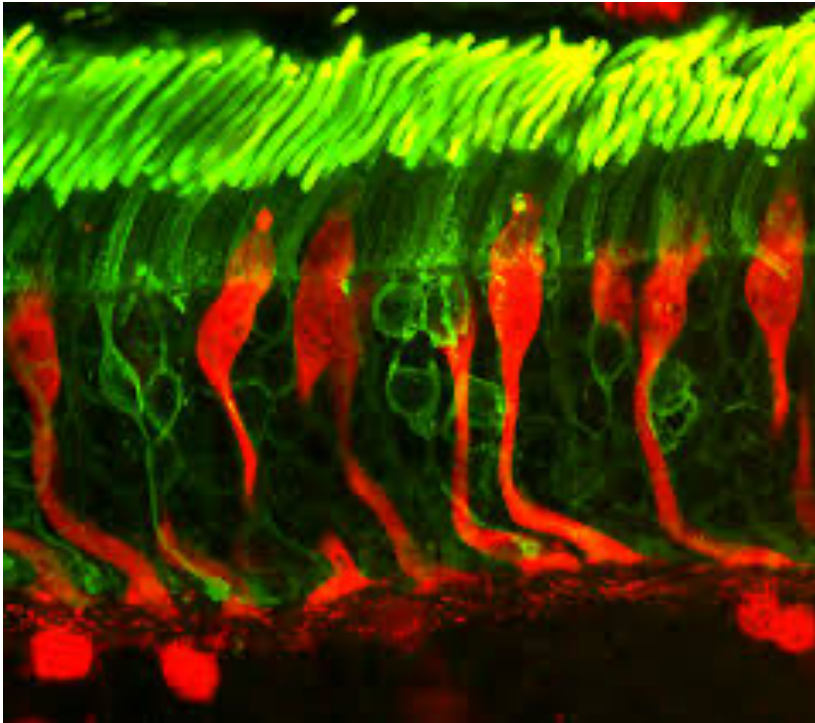
Luz e visão periférica (pouca nitidez) Campo de visão

Bastonetes –
maior concentração periférica

Cones –
maior concentração central



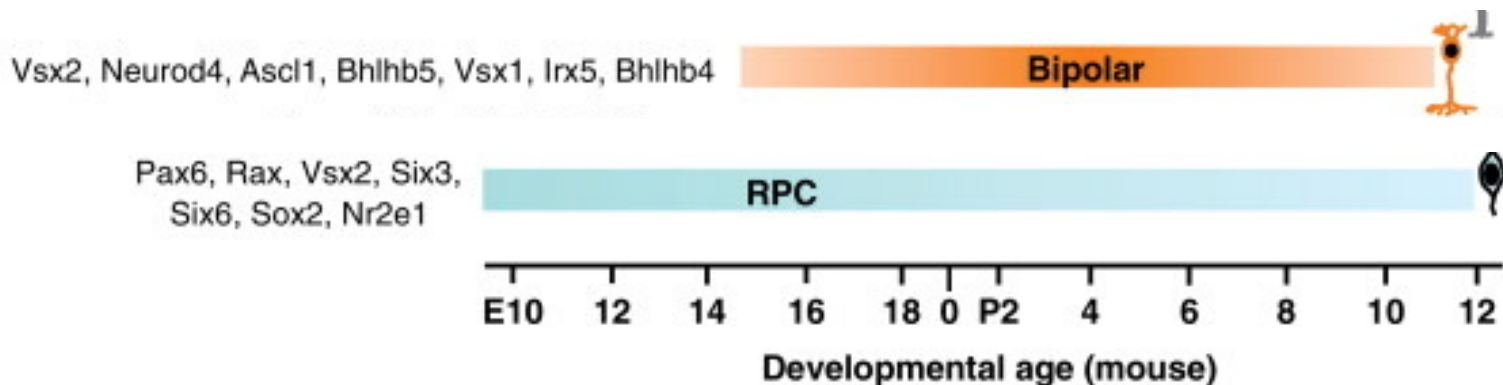
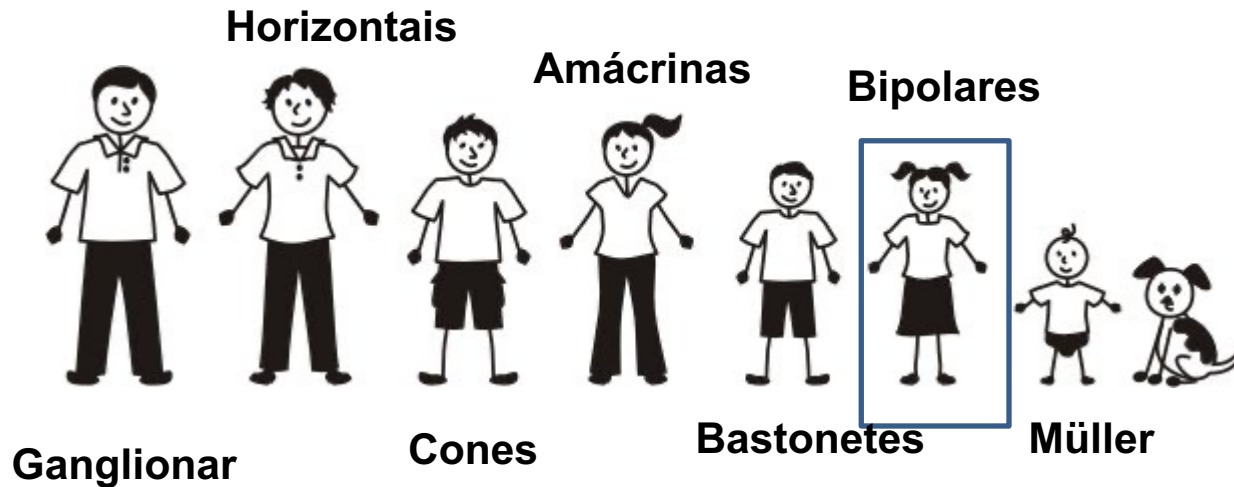


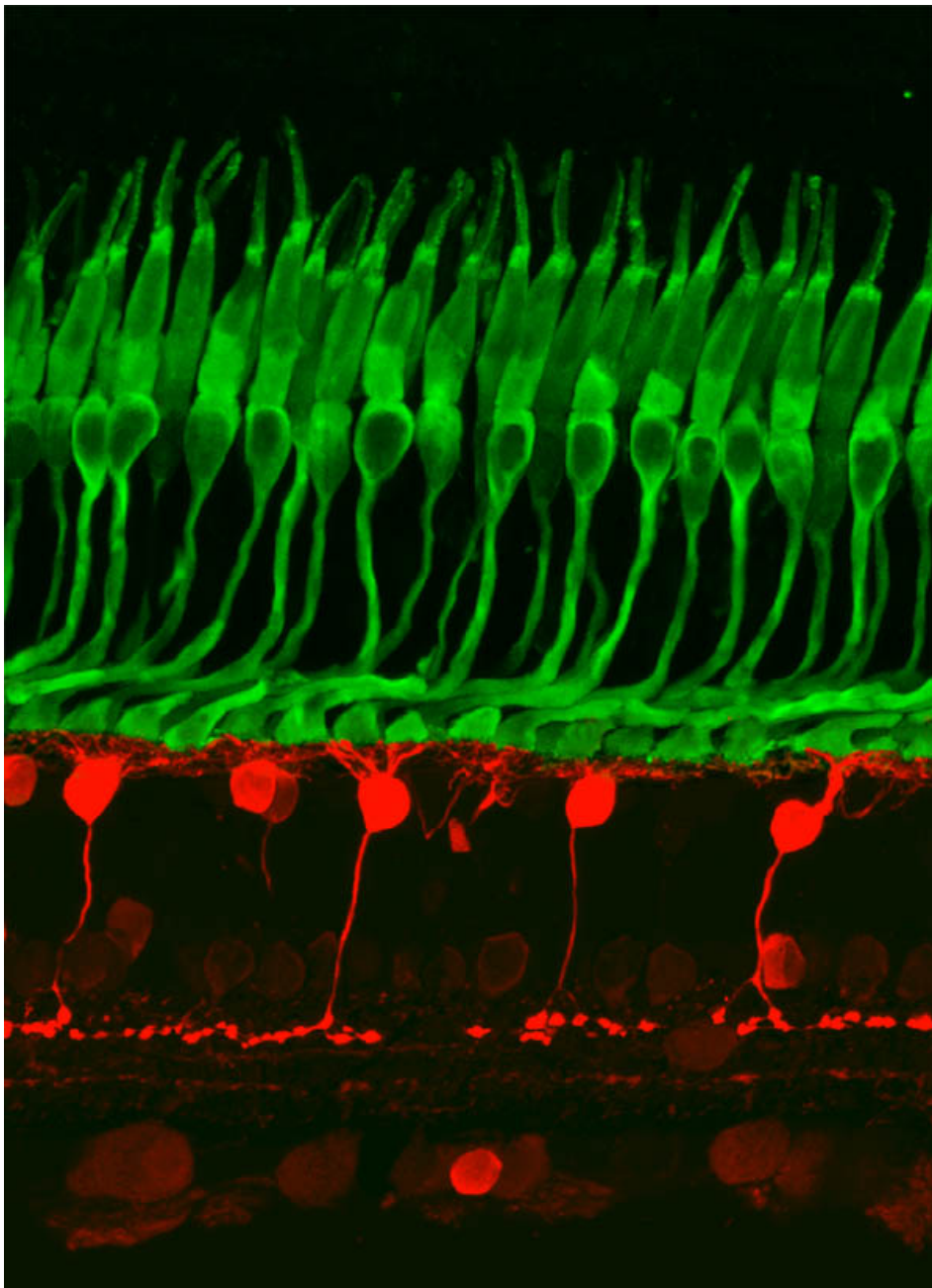


**Luz e visão periférica
(pouca nitidez)**

Diferenciação das células retinianas

Células Bipolares



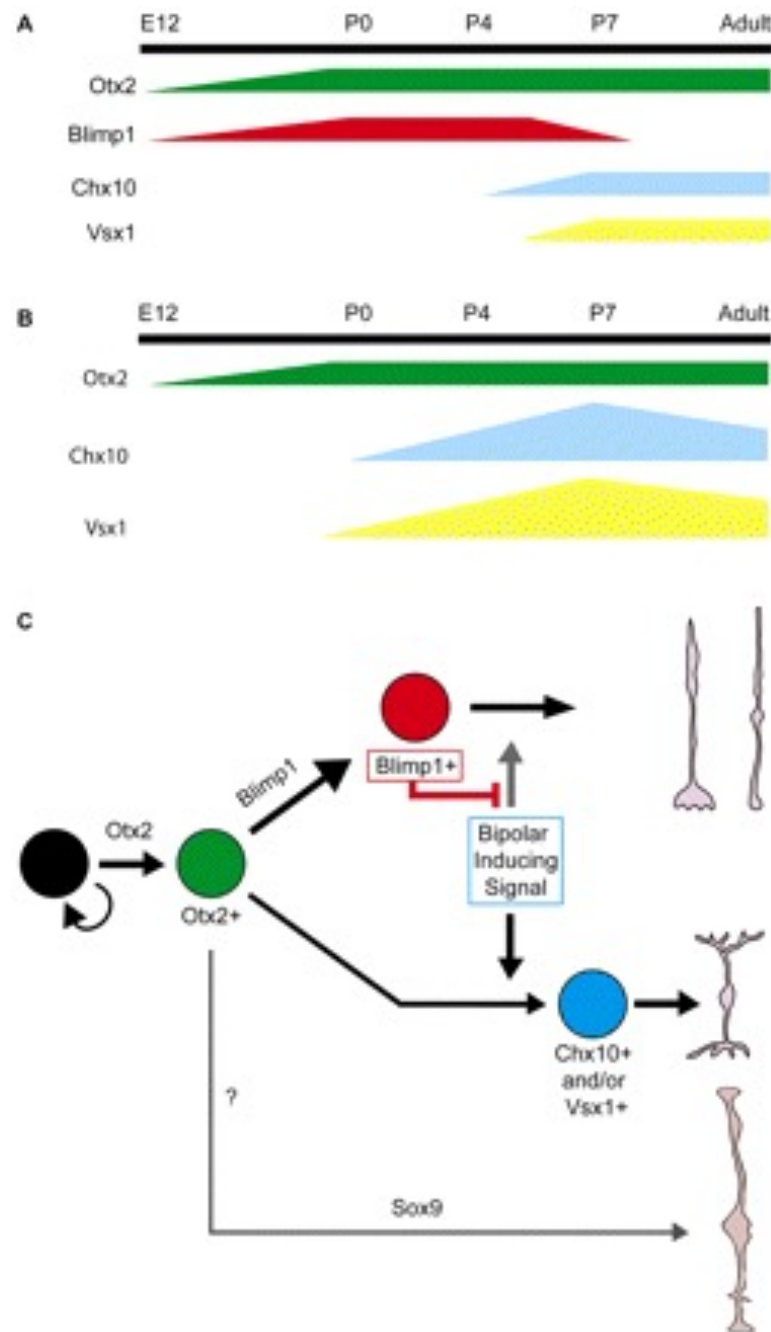


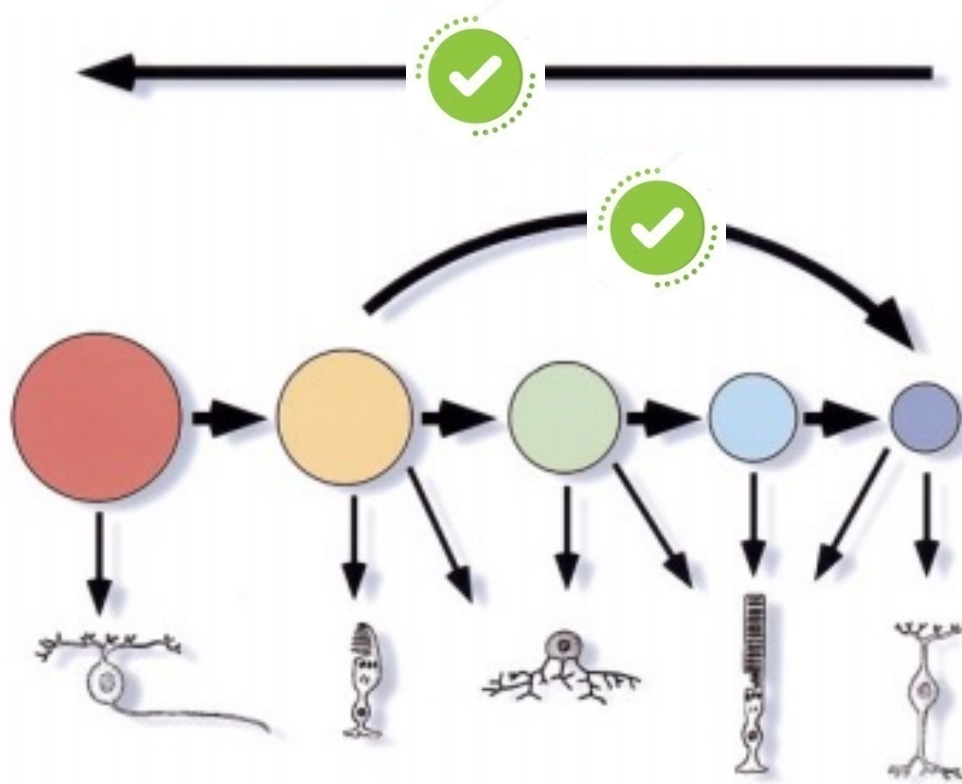
**Fazem sinapse com os
fotorreceptores**

- Bipolares ligadas à cones
- Bipolares ligadas à bastonetes

**Bipolares ON: despolarizam
Bipolares OFF: Hiperpolarizam**

**Refinam a informação “bruta” que é
transmitida pelo fotorreceptor e
modula informação espacia, temporal
e cromática**





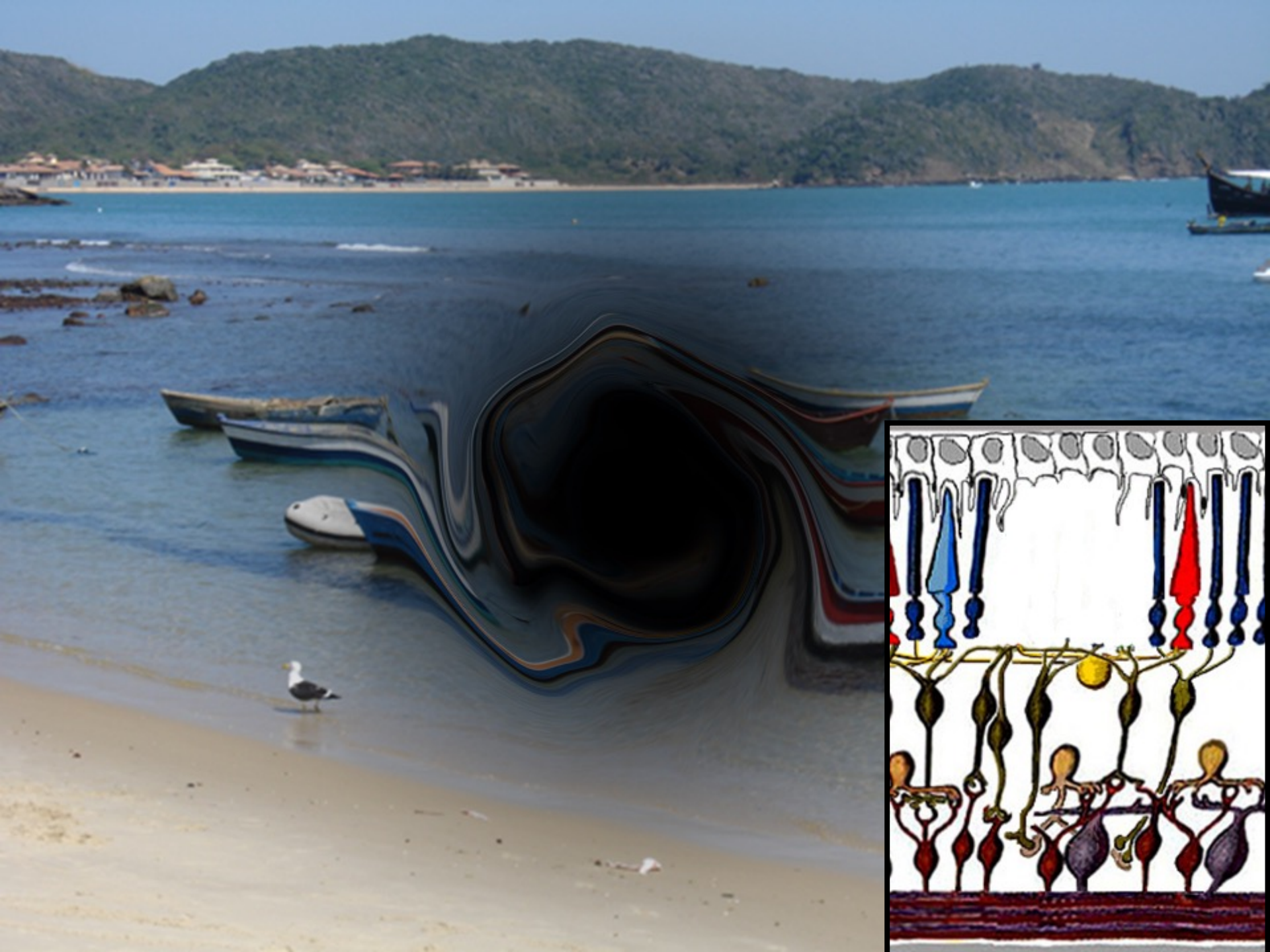
ENVIRONMENT

Aula: Desenvolvimento e Regeneração da Retina

Parte 4 - Regeneração Retiniana

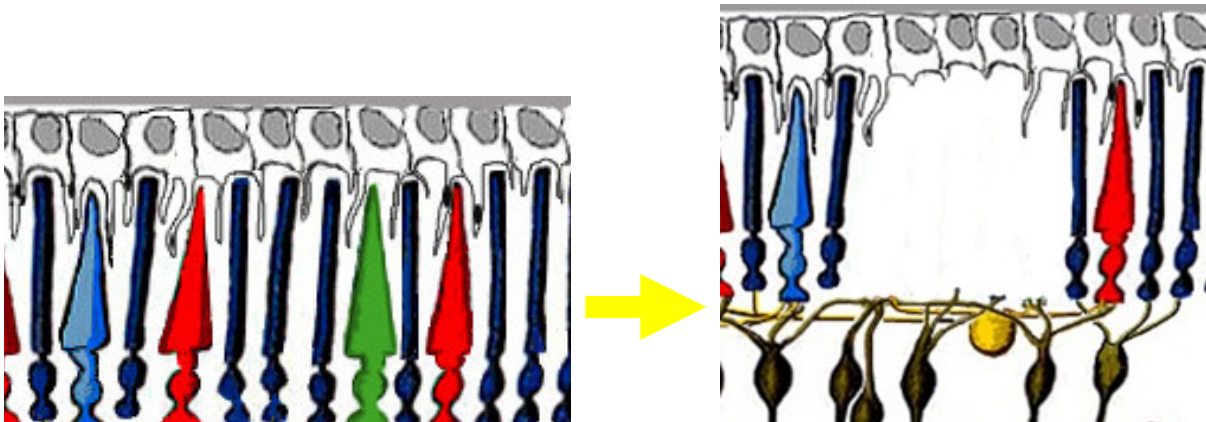






A morte dos fotorreceptores provoca perda visual progressiva e irreversível

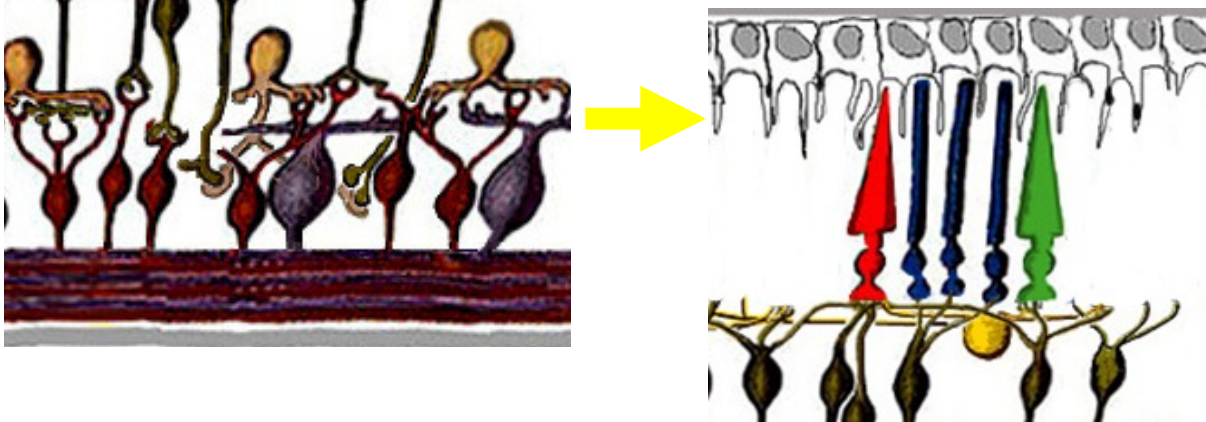
Degeneração central



Age Macular Degeneration



Degeneração periférica



Retinitis Pigmentosa



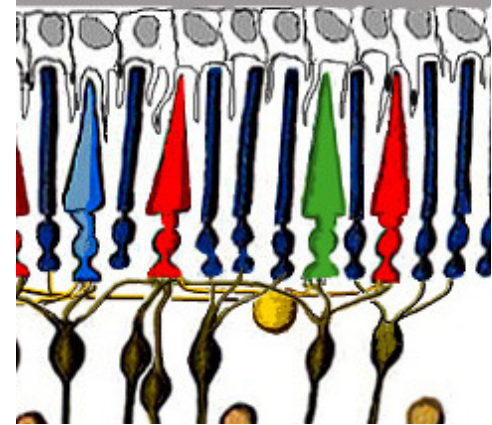
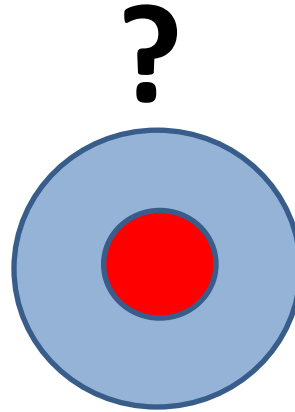
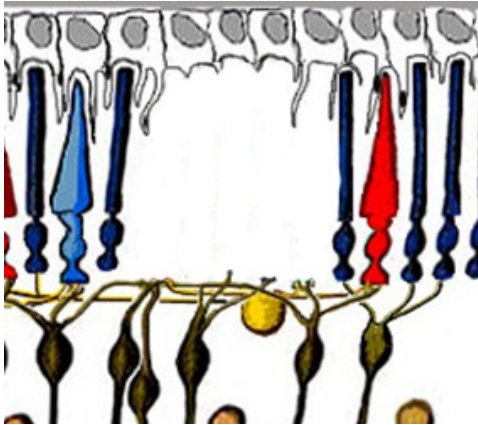


- ***Não existe tratamento efetivo***
- ***Não existe cura***

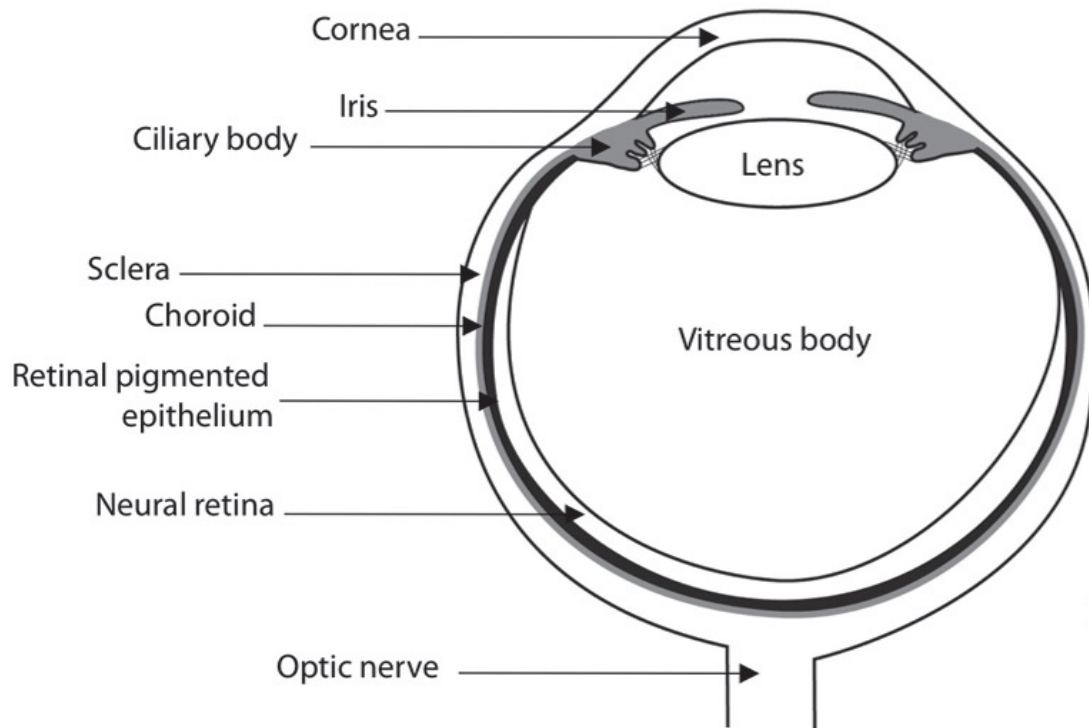
Tratamentos clínicos:

- ***Não reverterem o quadro degenerativo;***
- ***Não interrompem a degeneração;***
- ***Retardam o processo***

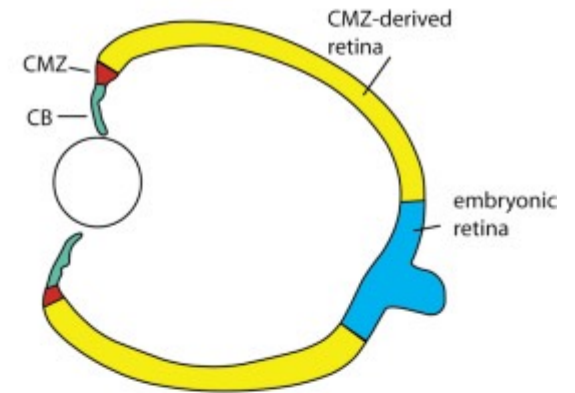
Células-tronco retinianas: esperança para a perda visual



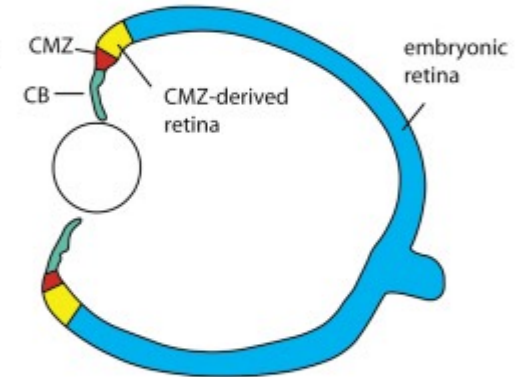
Regeneração da Retina



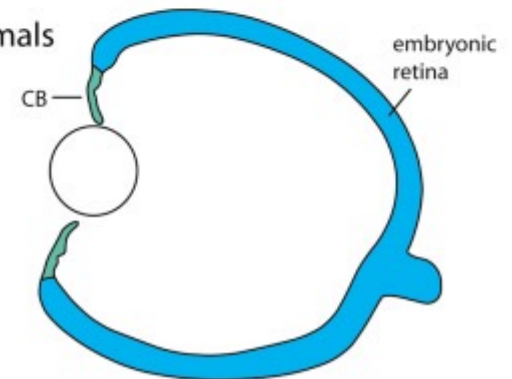
Frogs
and
Fish



Birds



Mammals

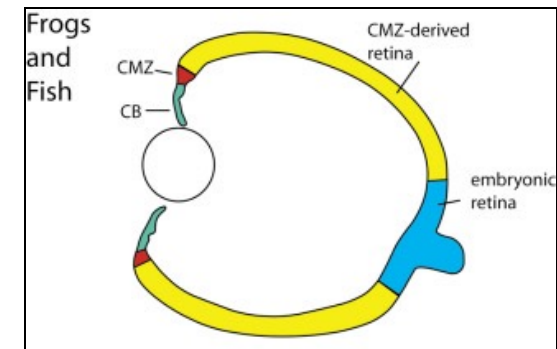
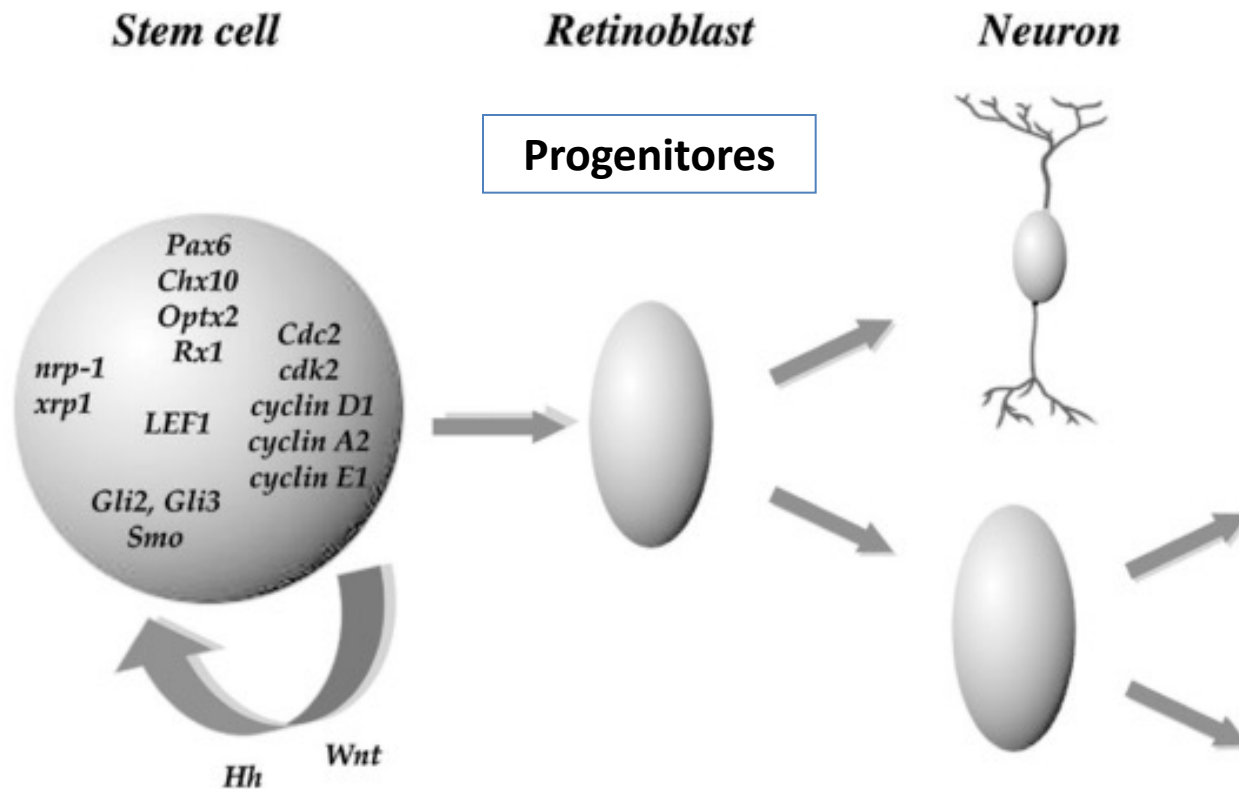






Epitélio Pigmentado Retiniano
 Células gliais de Müller
 Células do Epitélio Ciliar
 Células da Íris
 Células-tronco Residentes

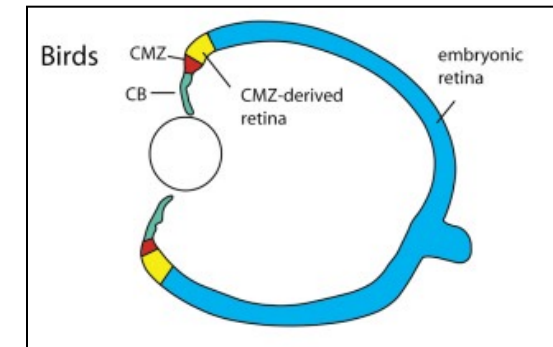
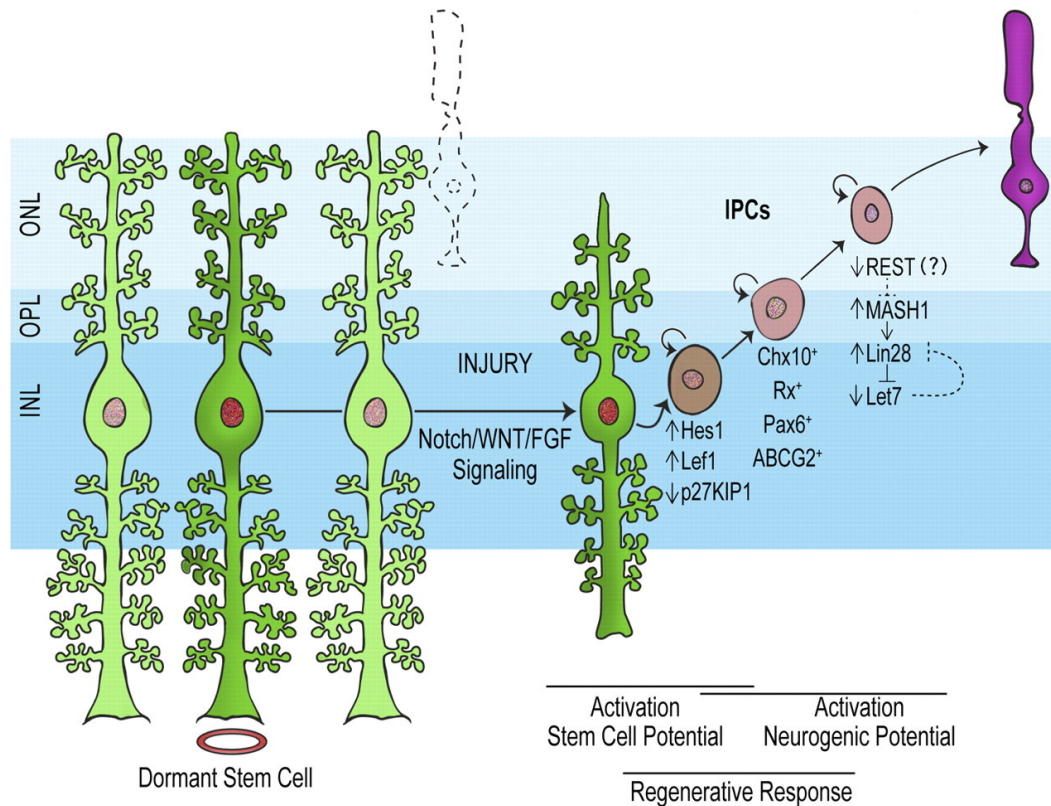
Reprogramação





Epitélio Pigmentado Retiniano
 Células gliais de Müller
 Células do Epitélio Ciliar
~~Células da Íris~~
~~Células-tronco Residentes~~

Reprogramação

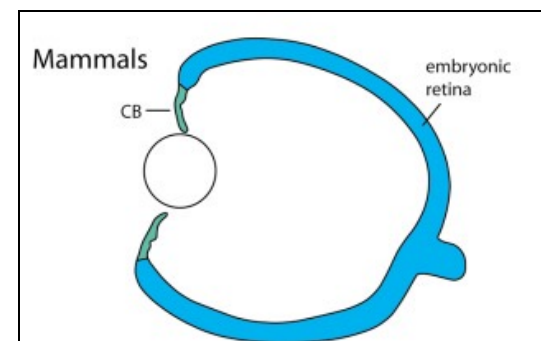
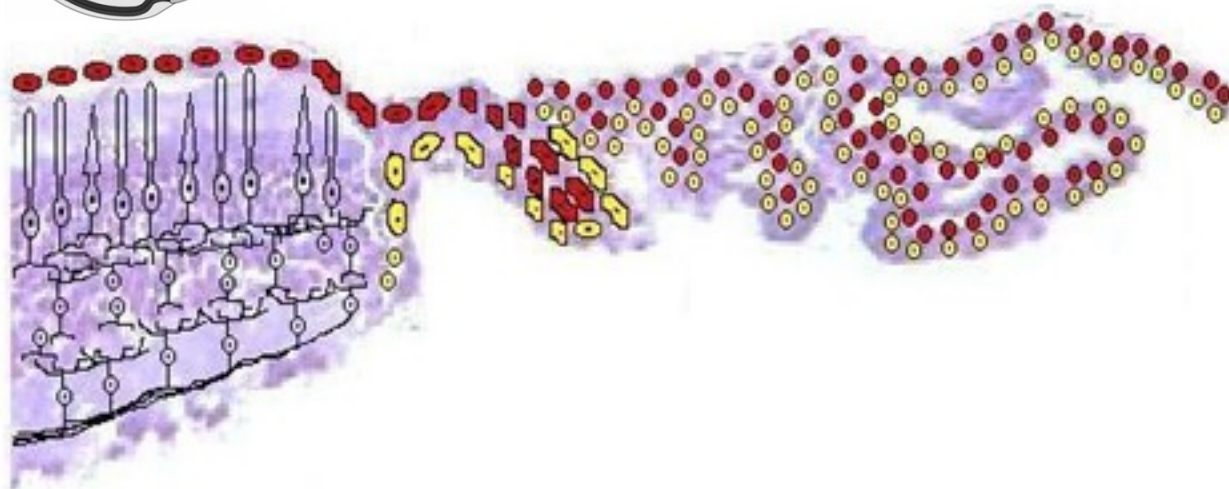
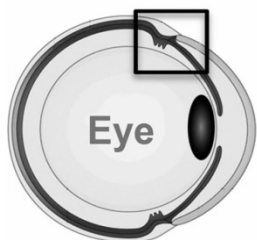


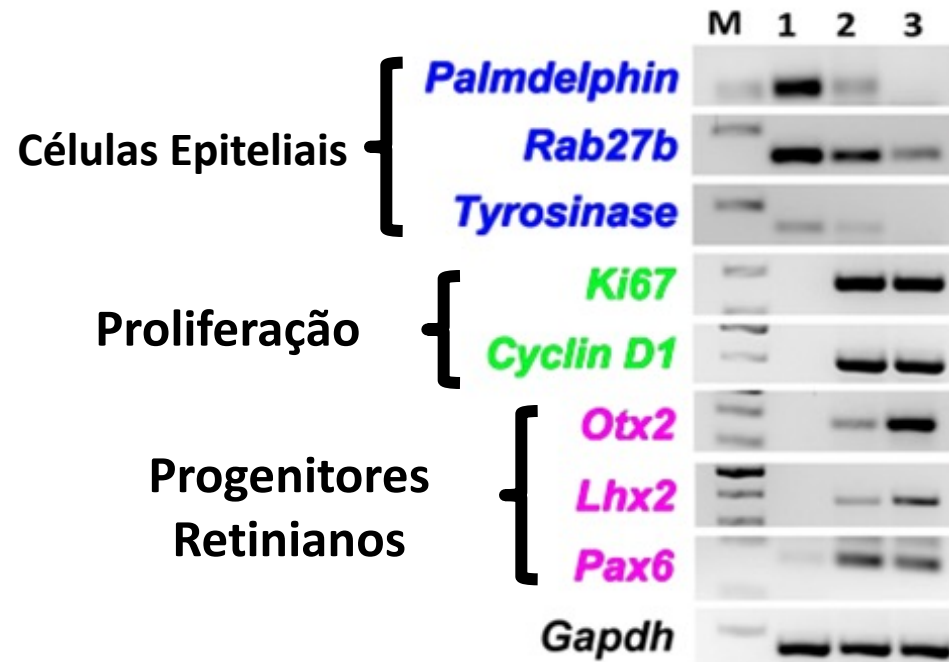
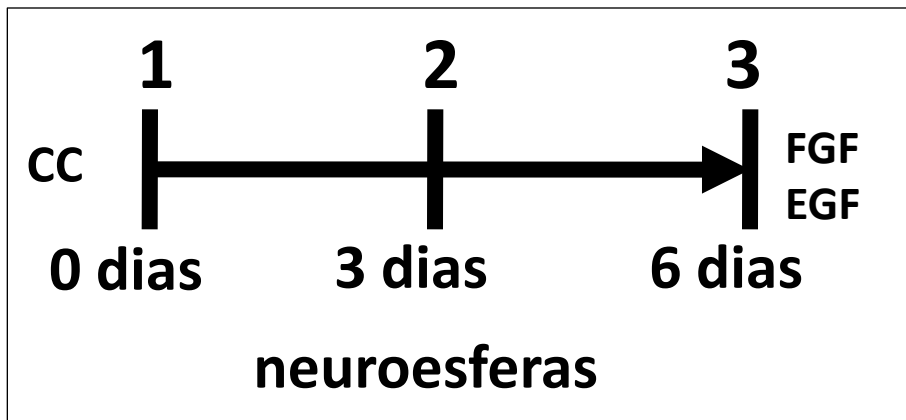
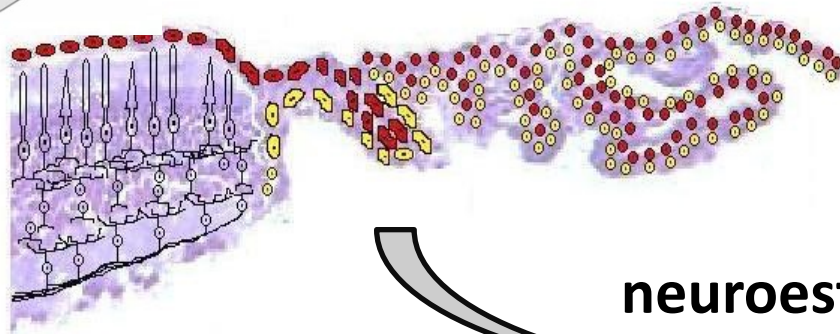


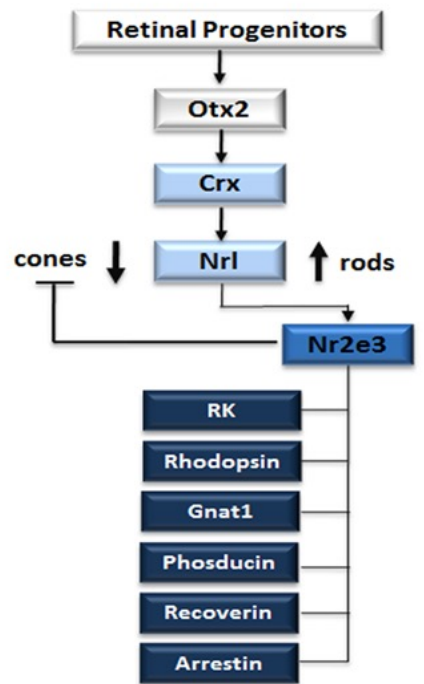
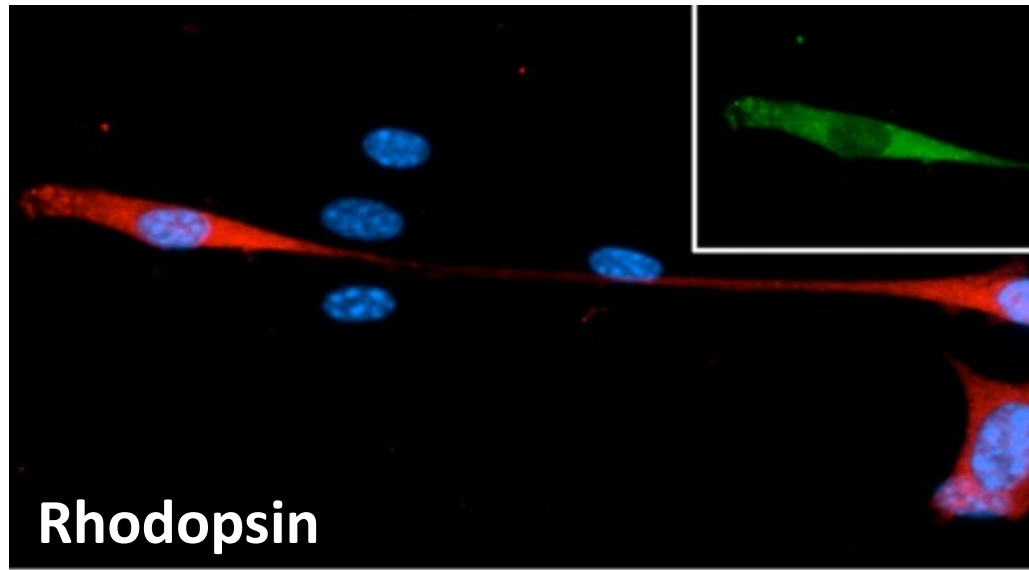
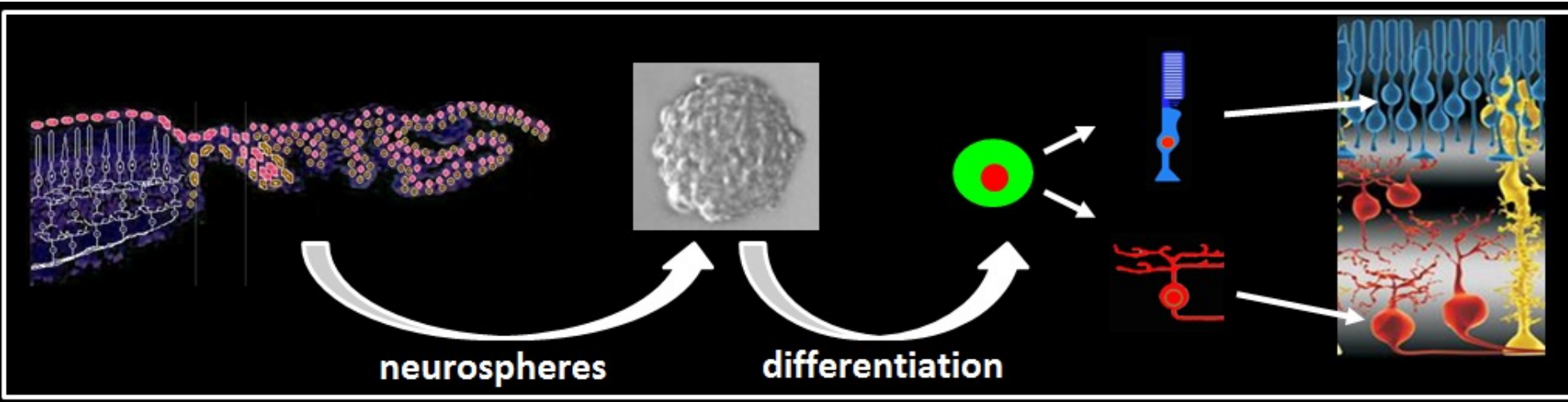
~~Epitélio Pigmentado Retiniano~~
~~Células gliais de Müller~~
~~Células do Epitélio Ciliar~~
~~Células da Íris~~
~~Células-tronco Residentes~~

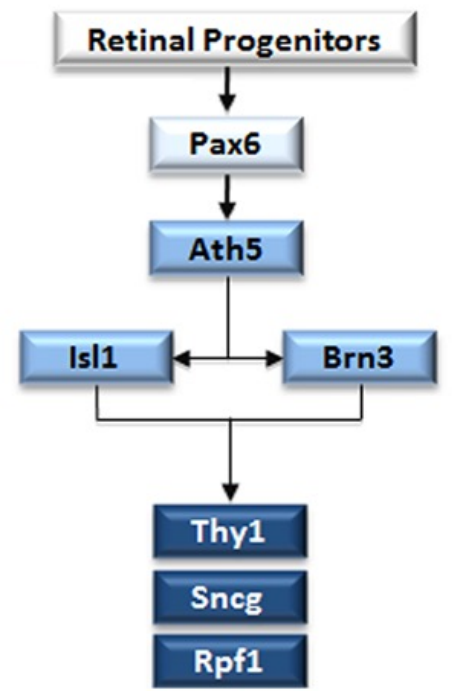
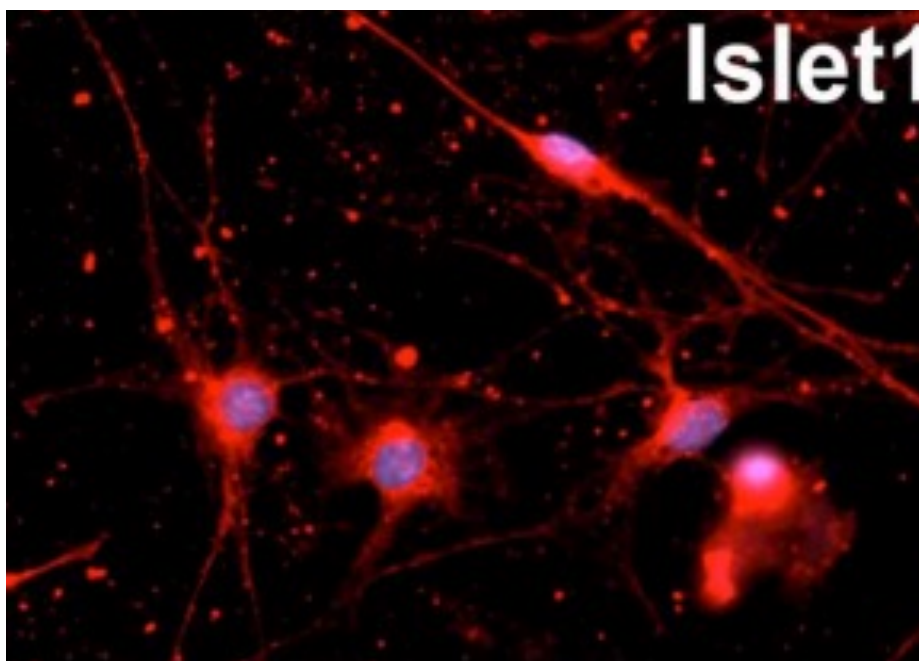
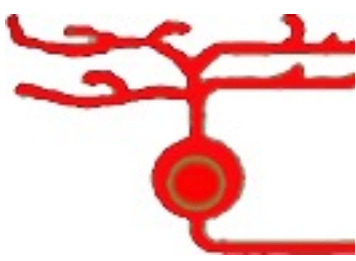
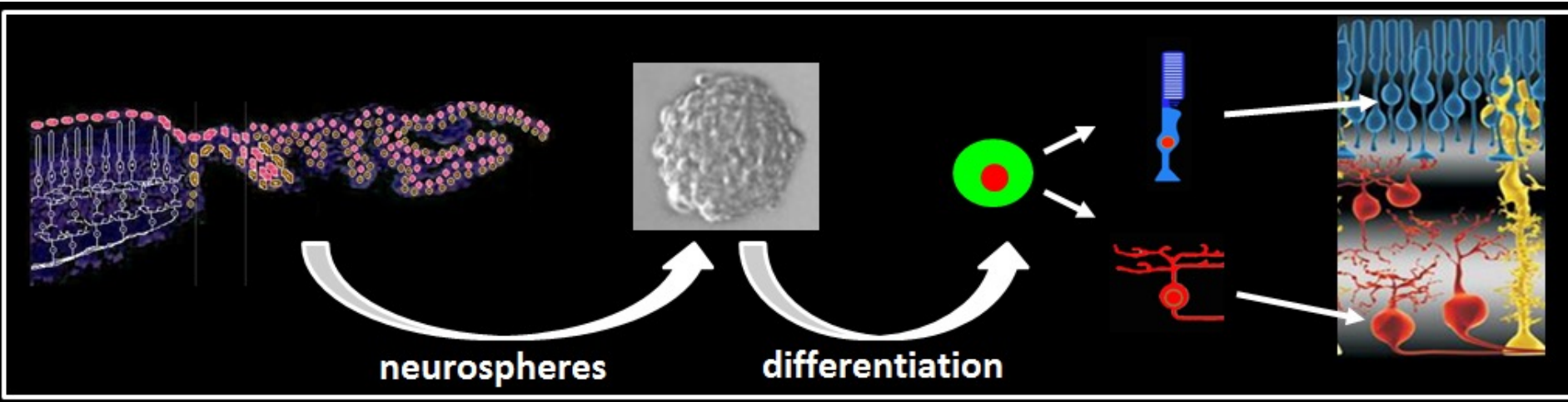
Células gliais de Müller
 Células do Epitélio Ciliar

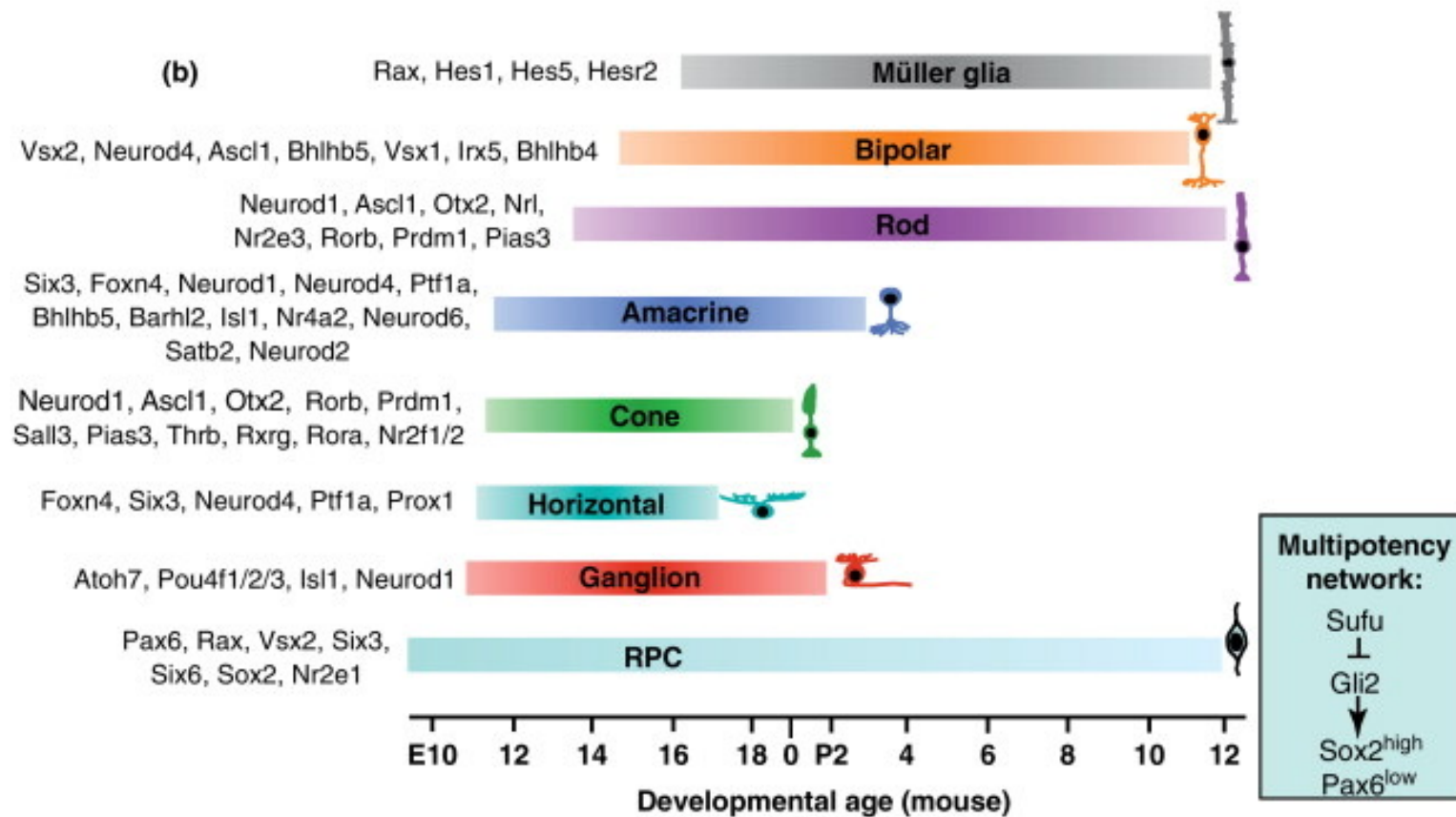
} **Indução de
Reprogramação**



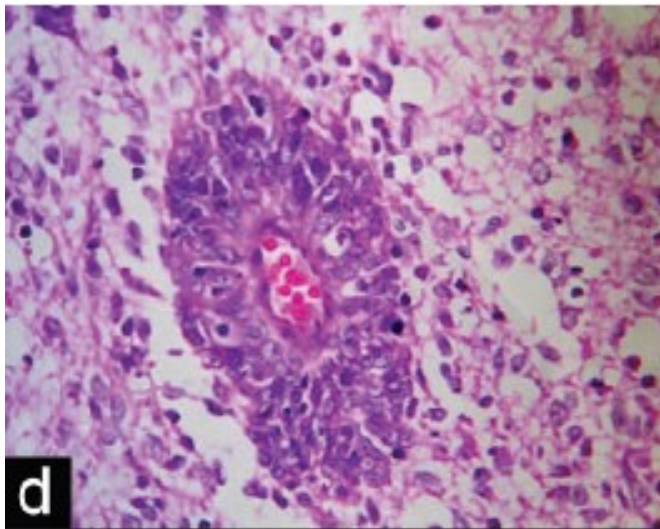




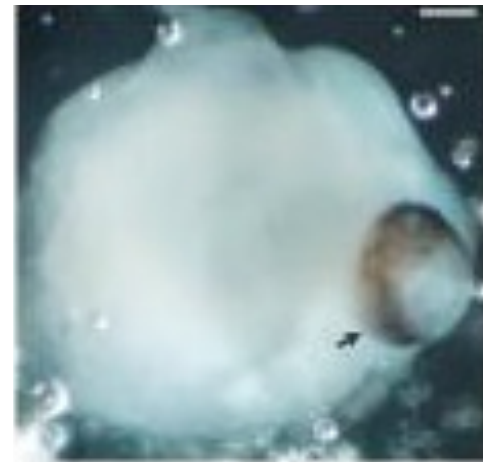
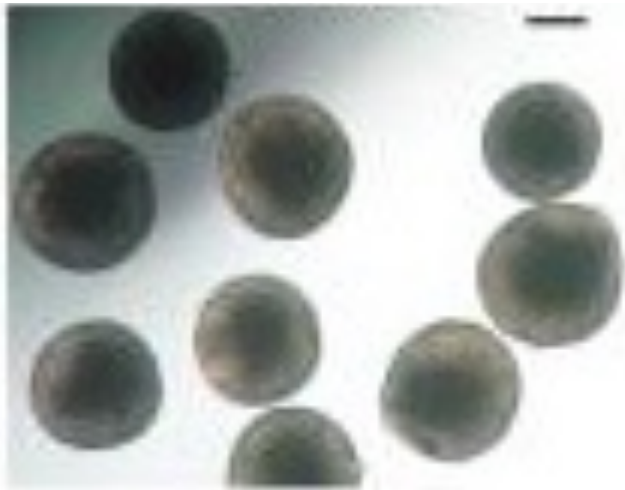
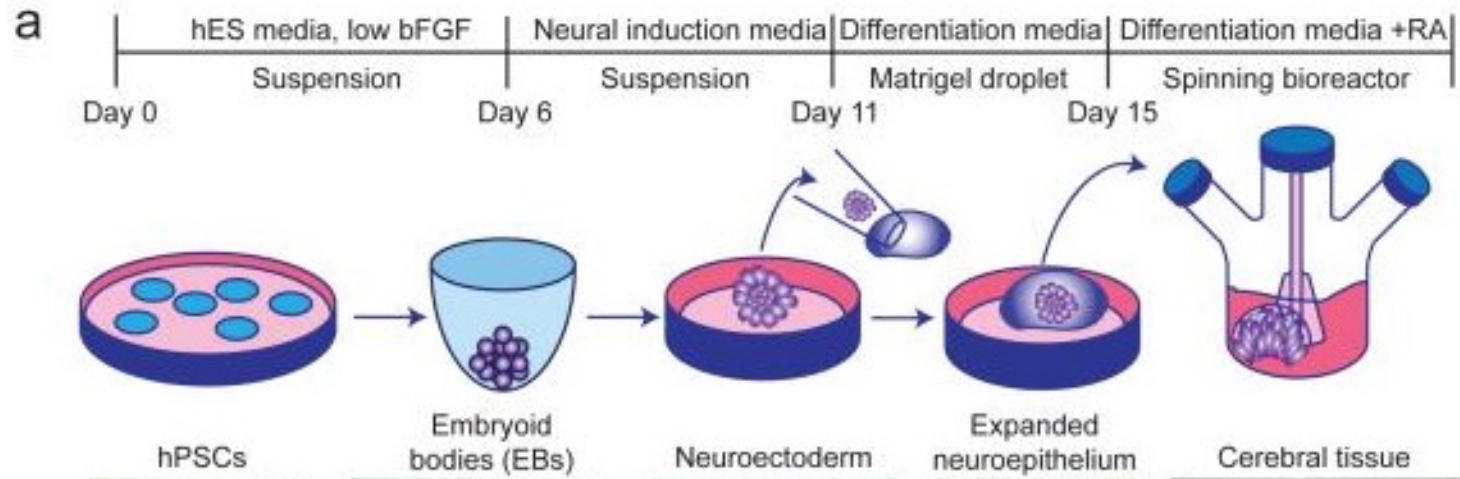


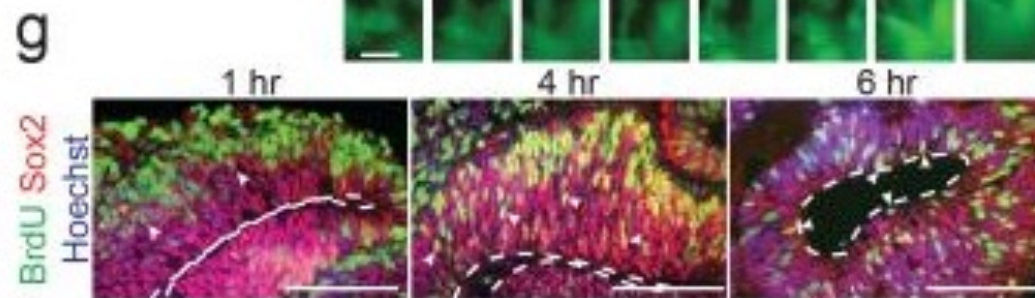
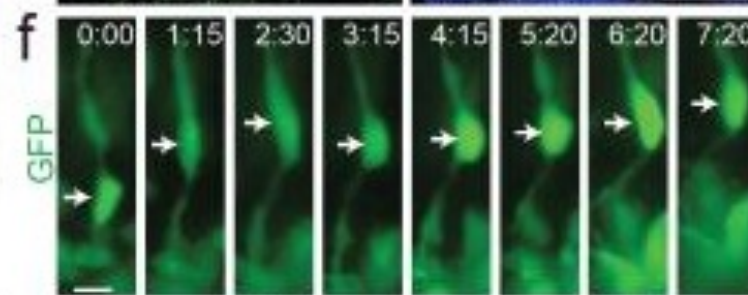
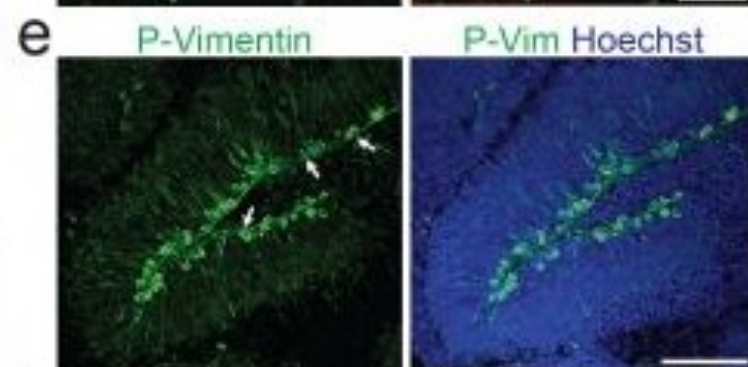
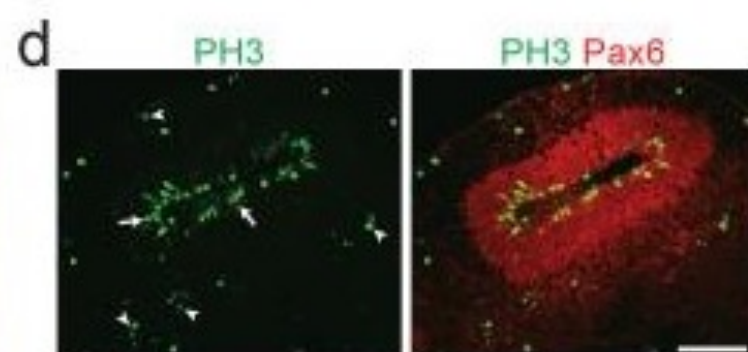
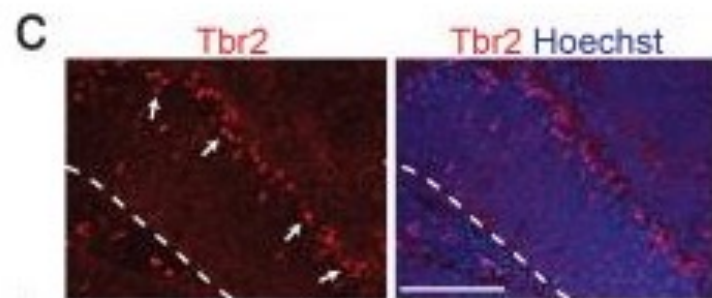
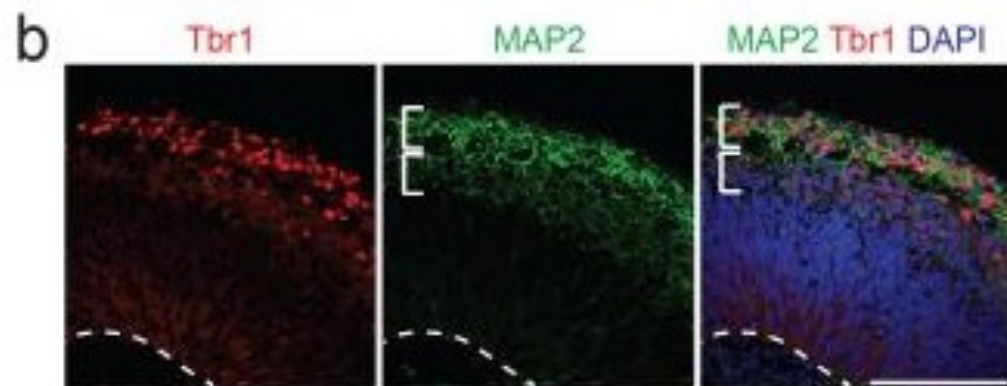
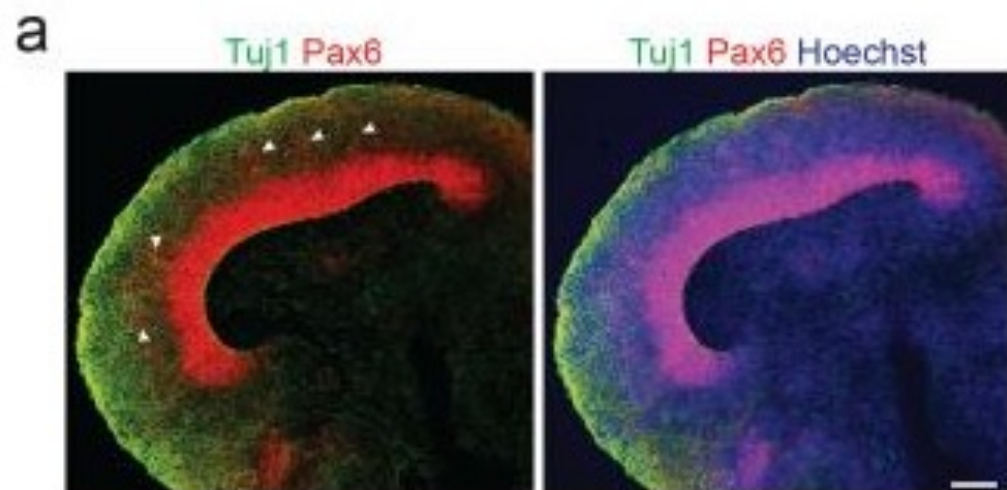


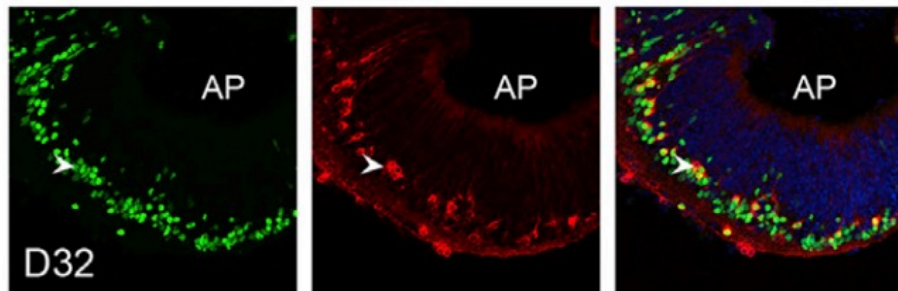
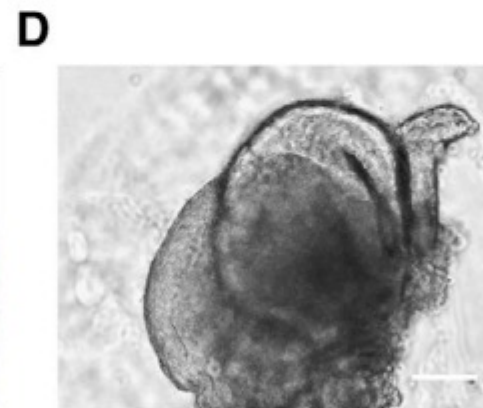
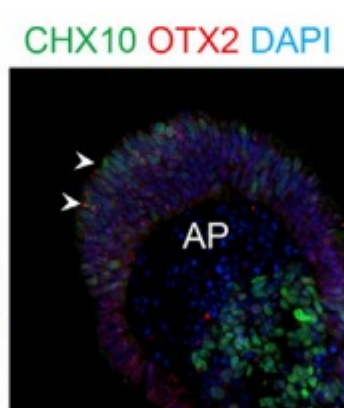
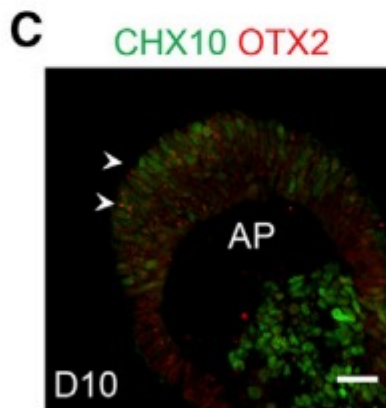
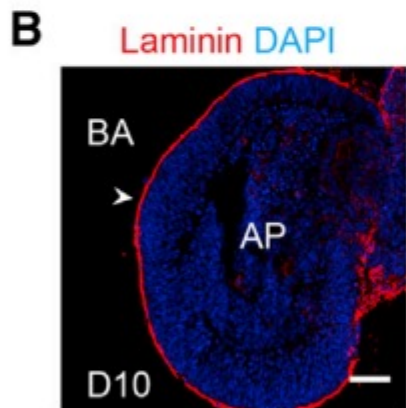
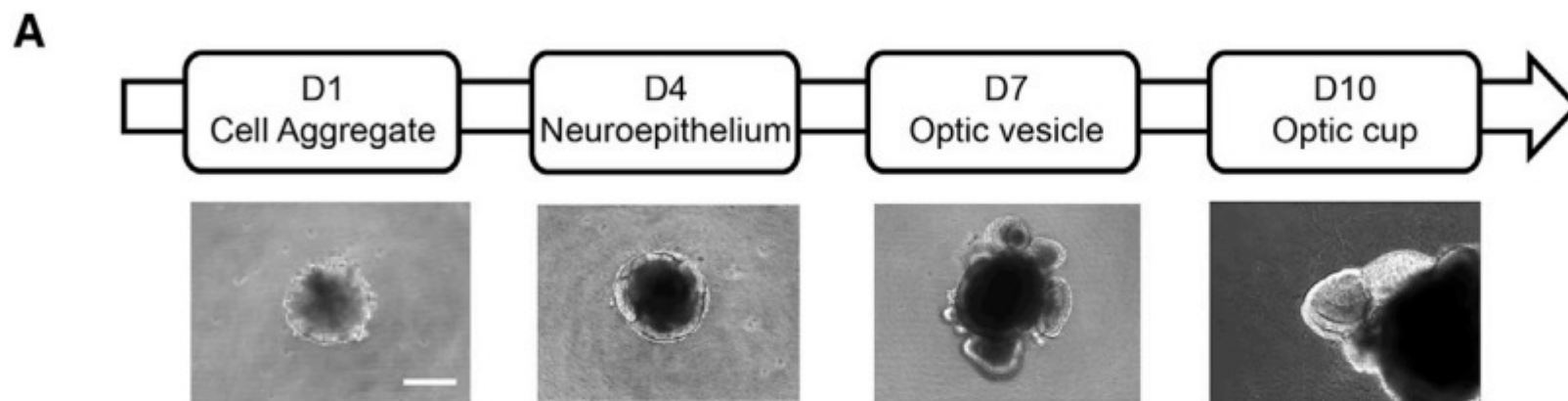
Mini-brain ou cultura organóide de cérebro



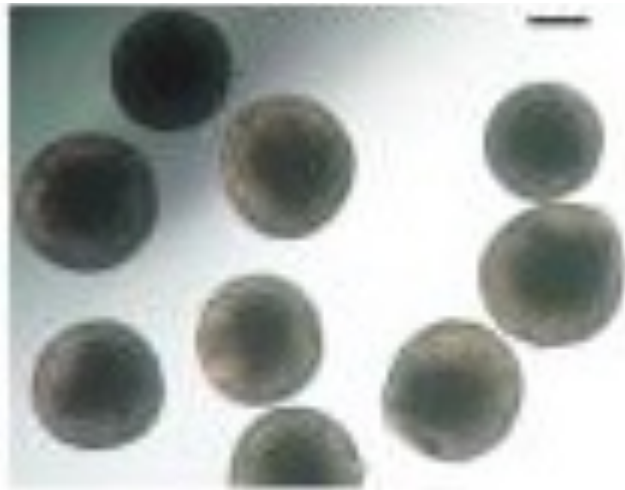
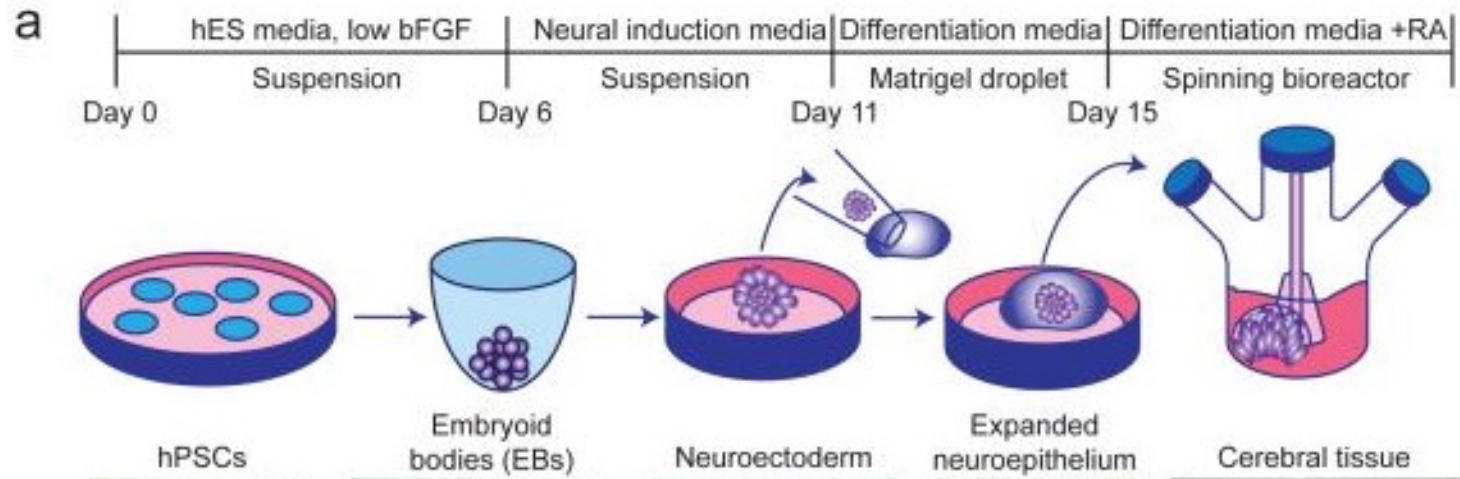
Rosetas = estruturas esféricas onde as células da região central proliferam e as da periferia se diferenciam em neurônios







CHX10 (green) PKCα (red) DAPI (blue)



C

Sox2 Tuj1 Hoechst

