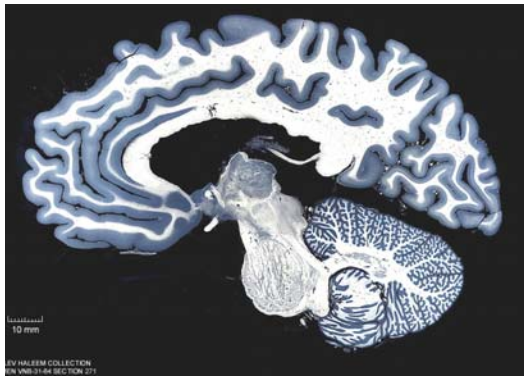
Evolution of the animal central nervous system: From *Platynereis* to vertebrates

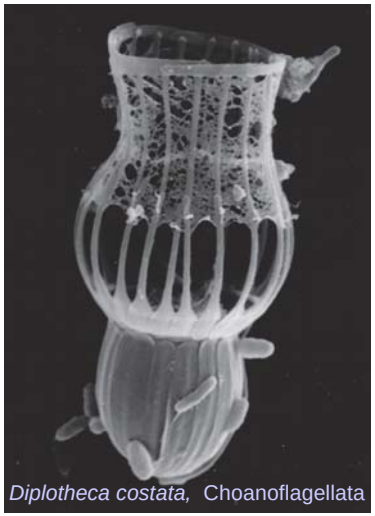
April 23rd, 2008

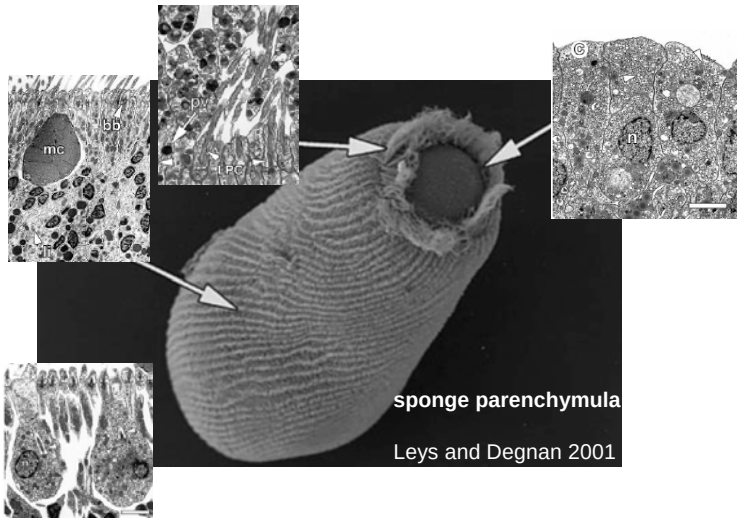
KITP brain'08

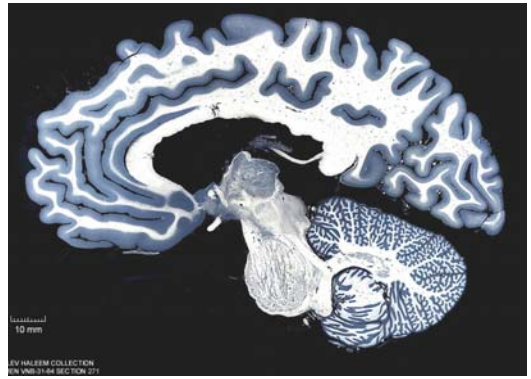


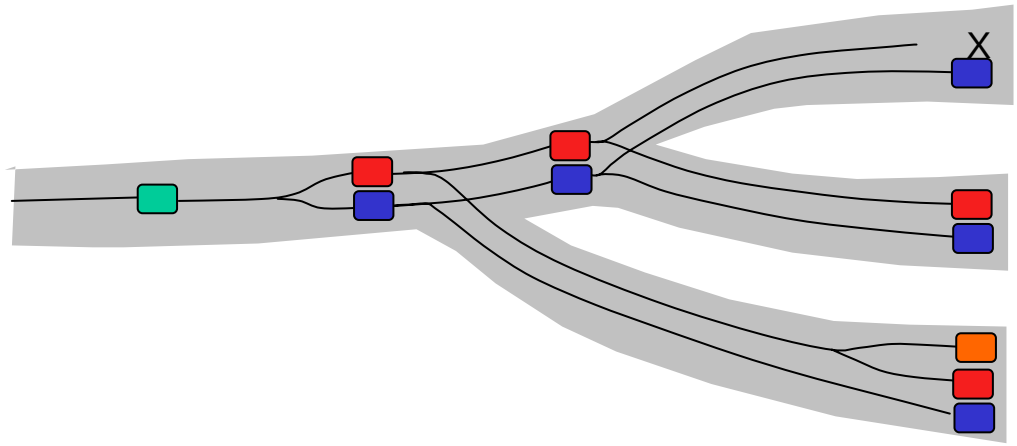
Homology research: comparative embryology, cytology and anatomy
(There is no other way to unravel body plan evolution and CNS evolution.)

Cell type diversification


Diplotheca costata, Choanoflagellata







homologous cell types

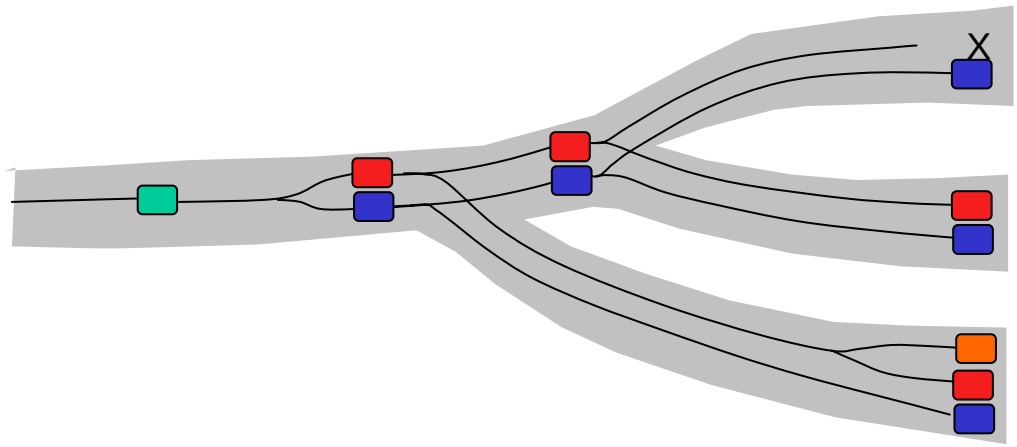
sister cell types

Comparative analysis of cell types

- transcription factor signature
- differentiation gene signature

  → physiology, morphology

molecular fingerprint



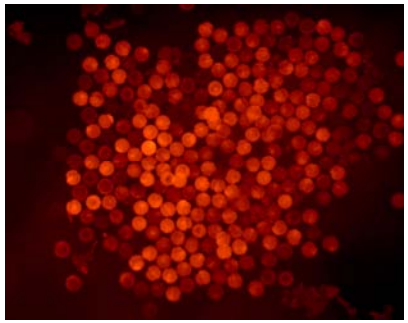
homologous cell types

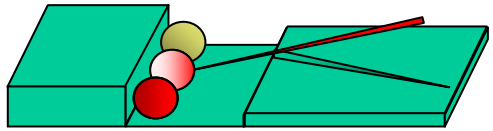
sister cell types

Platynereis dumerilii

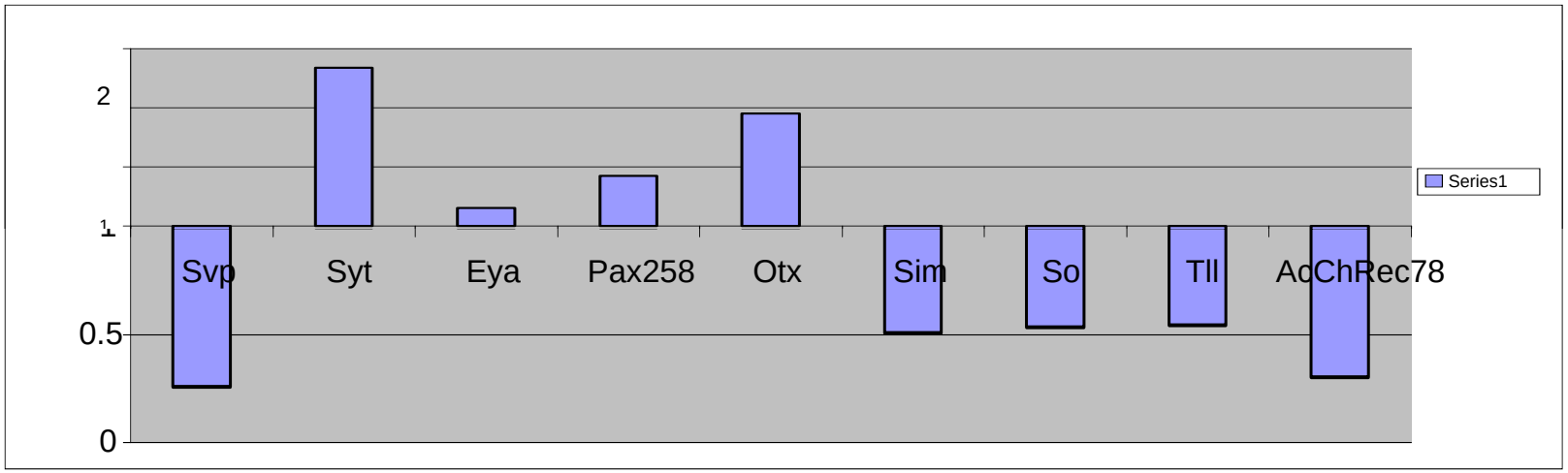
Platynereis dumerilii
Phyllodocida
Annelida
Lophotrochozoa

Up- and downgregulation of genes in pax6 morphants





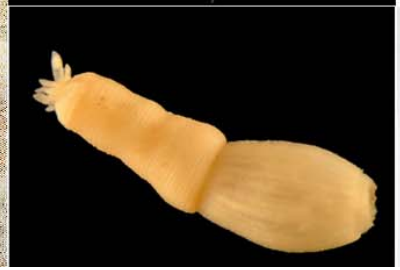
Benjamin Backfisch
Nicola Kegel
Raju Tomer



The search for Urbilateria: living fossils

after S. Conway Morris

Platynereis



priapulid

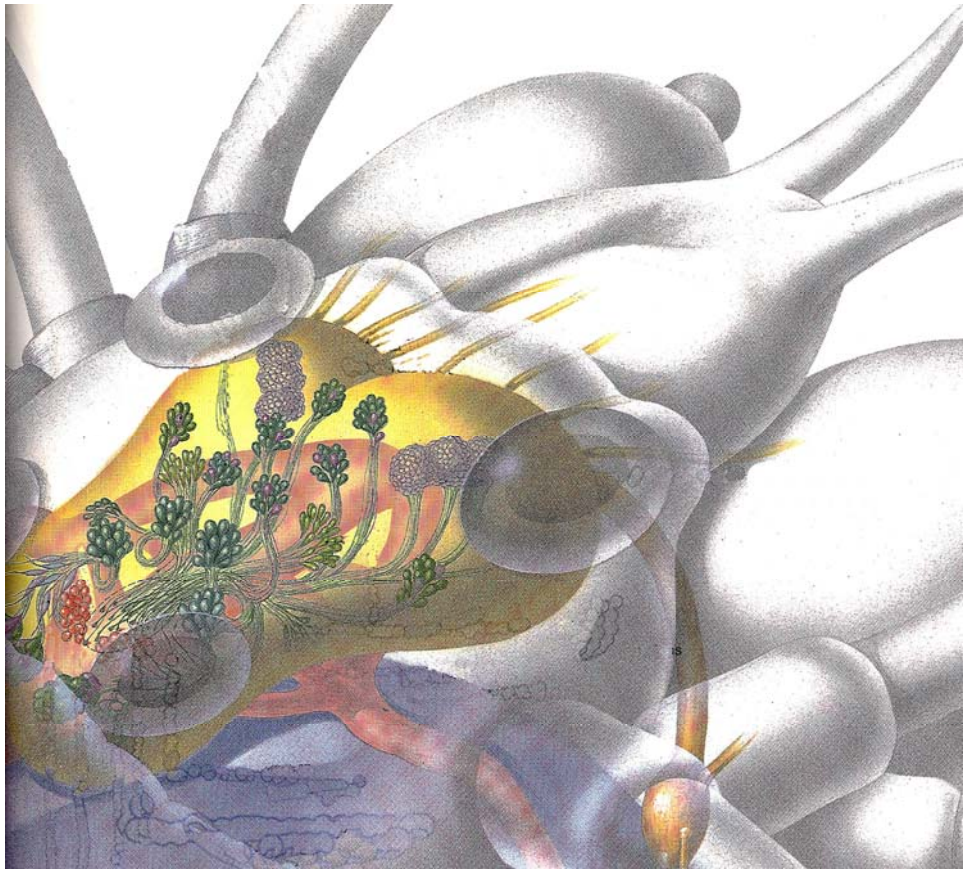
brachiopod

L:imulus

polyplacophoran

onychophoran

The *Platynereis* cerebral ganglia



Vasotocin

GnRH

POMC

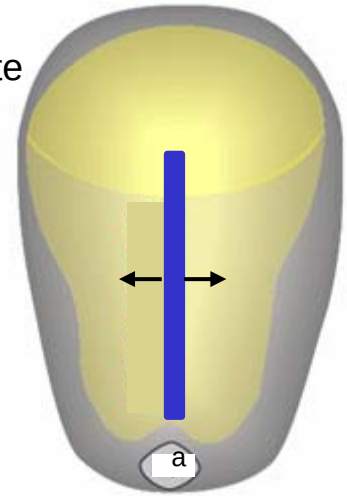
…

vertebrate pharmacology works!

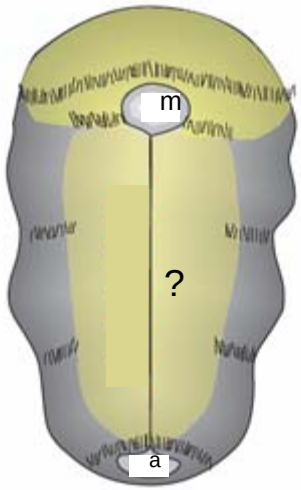
Matsumoto, A. and Ishii, S 1992

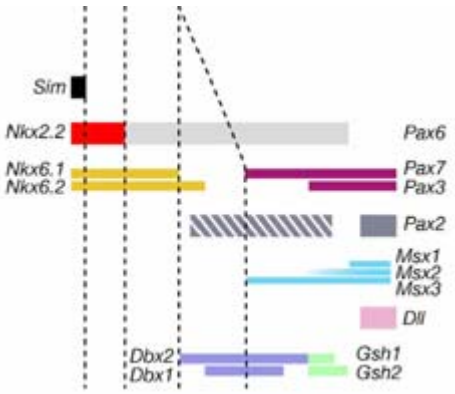
Mediolateral patterning in the trunk neuroectoderm

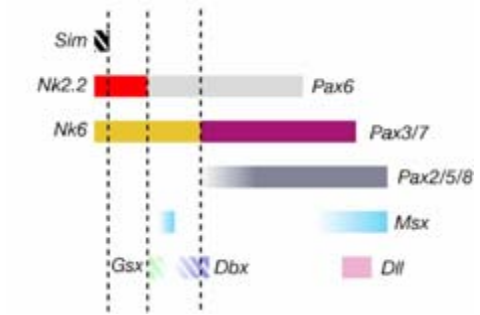
vertebrate



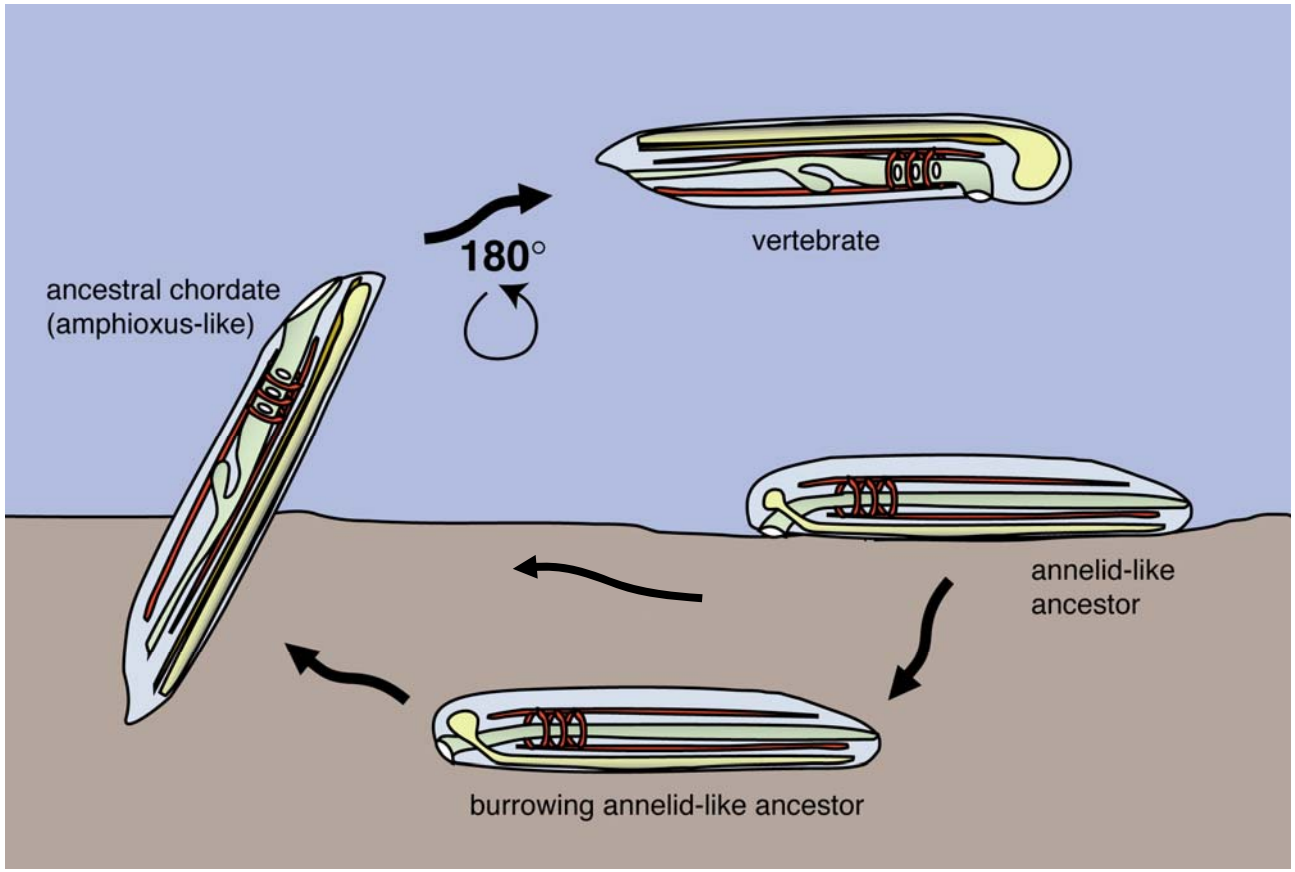
Platynereis



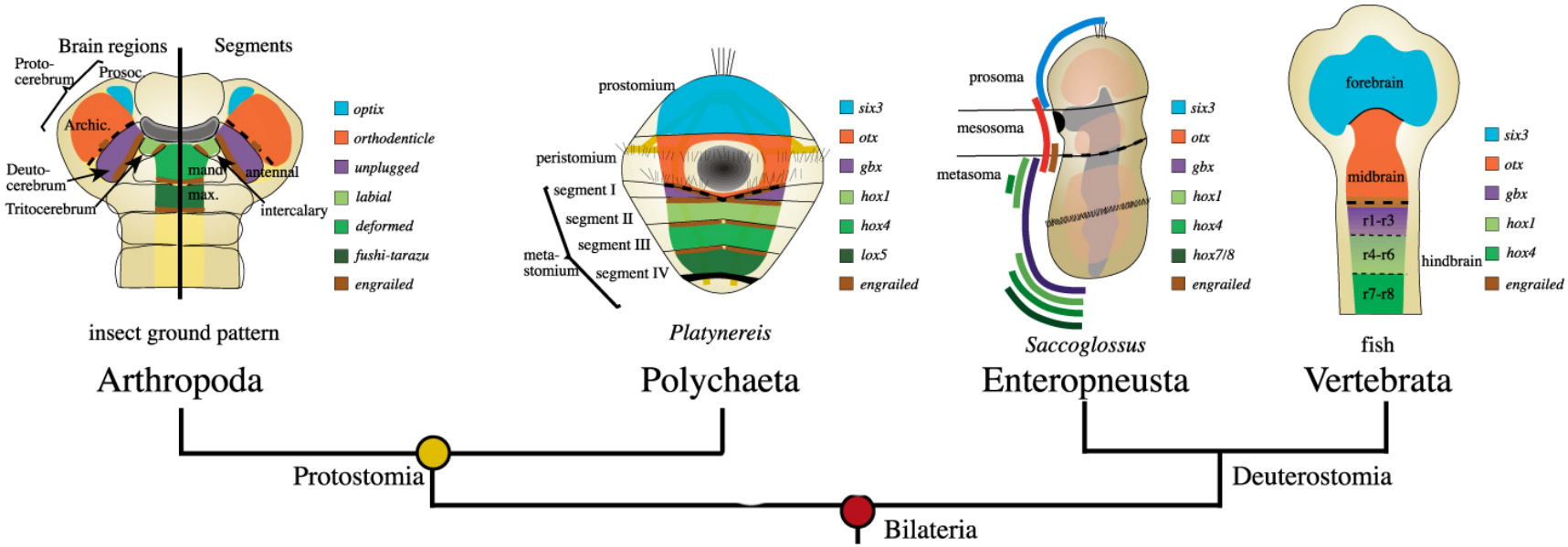




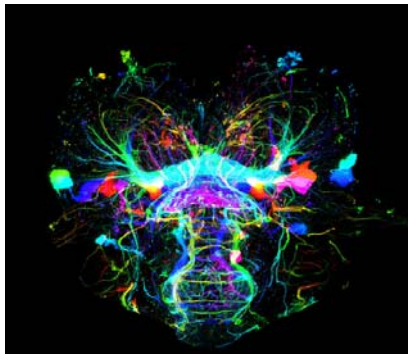
DV axis inversion (G. St. Hilaire, A. Dohrn)



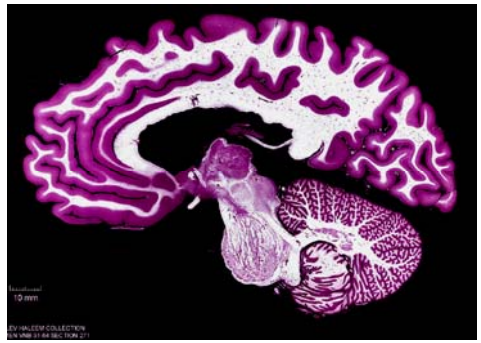
Anteroposterior patterning of the neuroectoderm



(collaboration with Gregor Bucher, Roman Kostyuchenko and Michael Akam)



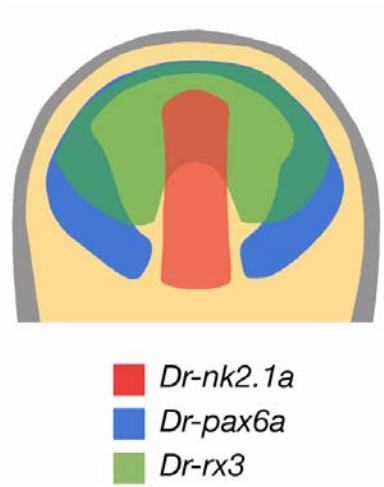
Evolution of neurosecretory brain centres



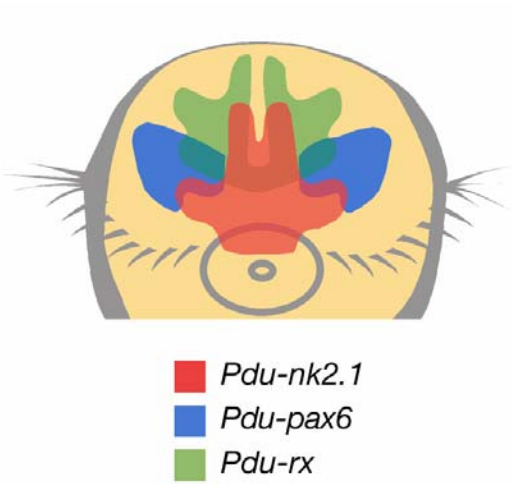
Kristin Tessmar-Raible
Cell **129**, 1389–1400 (2007)

Forebrain regionalisation in *Platynereis* and fish

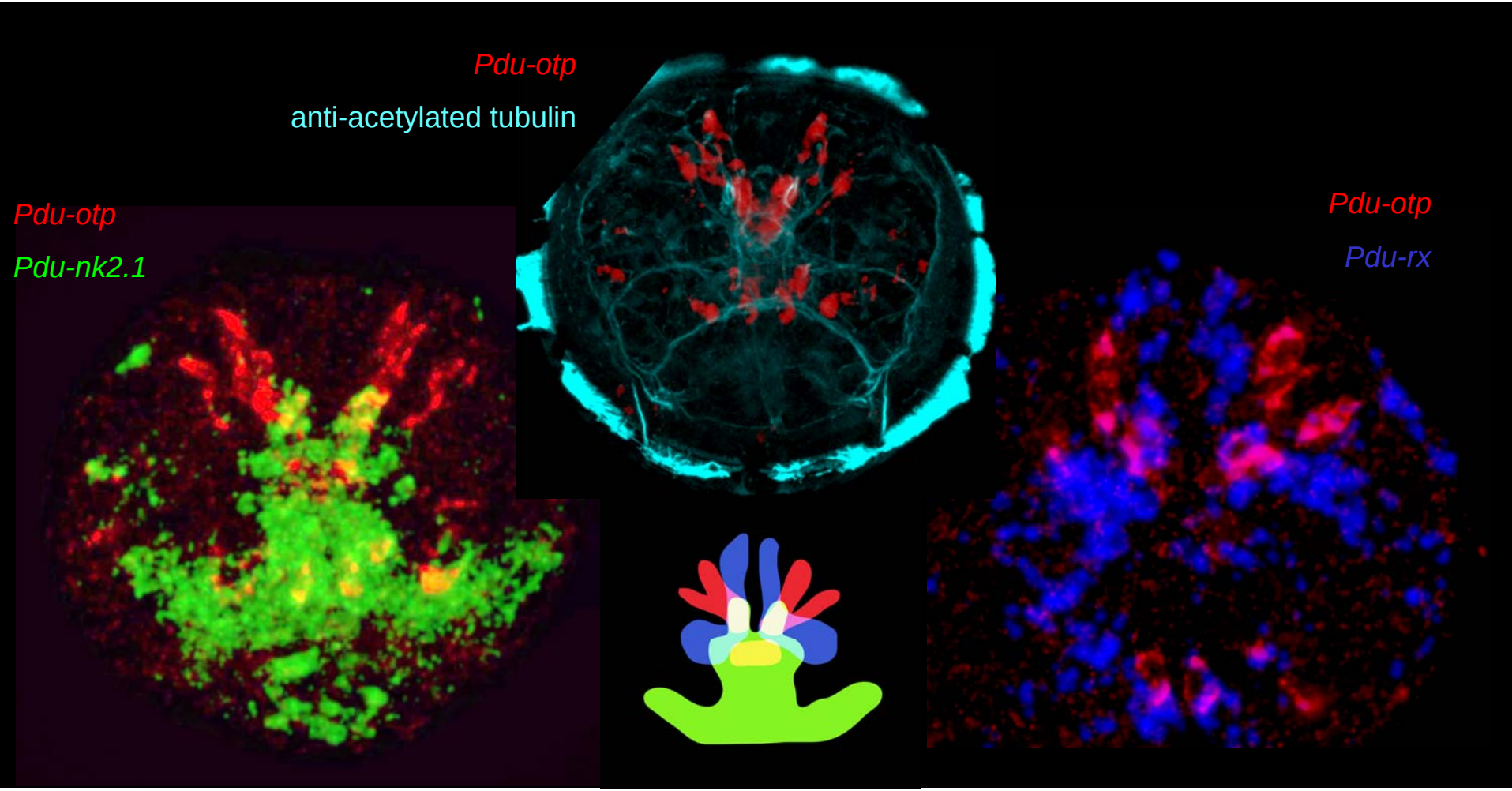
fish



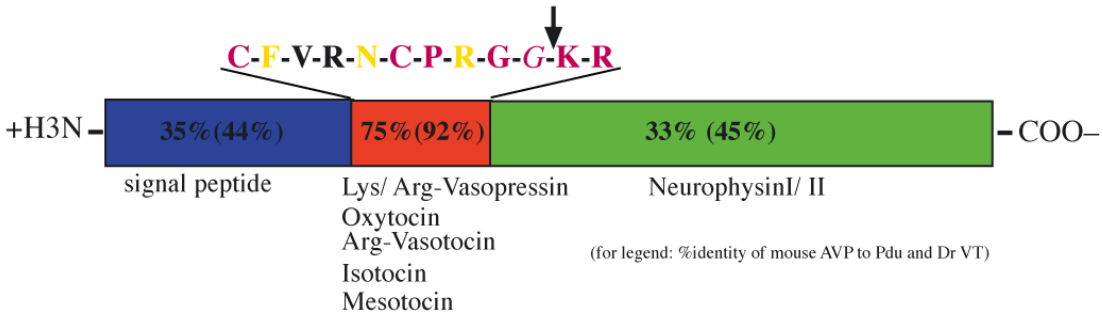
Platynereis

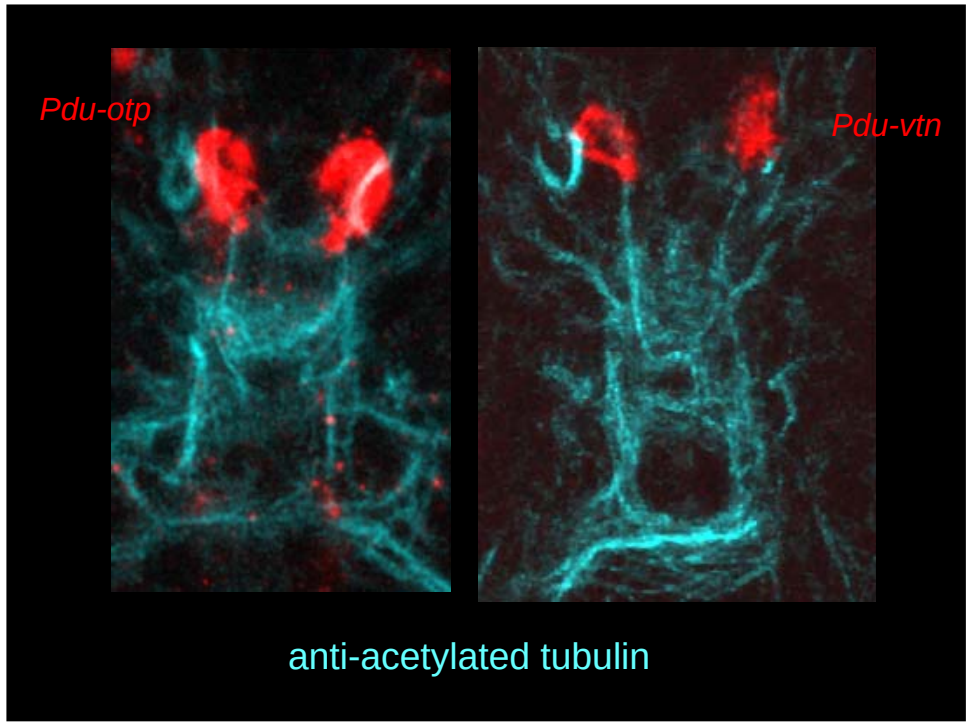


A small set of *nk2.1*+, *rx*+ and *otp*+ cells in 48hpf *Platynereis*

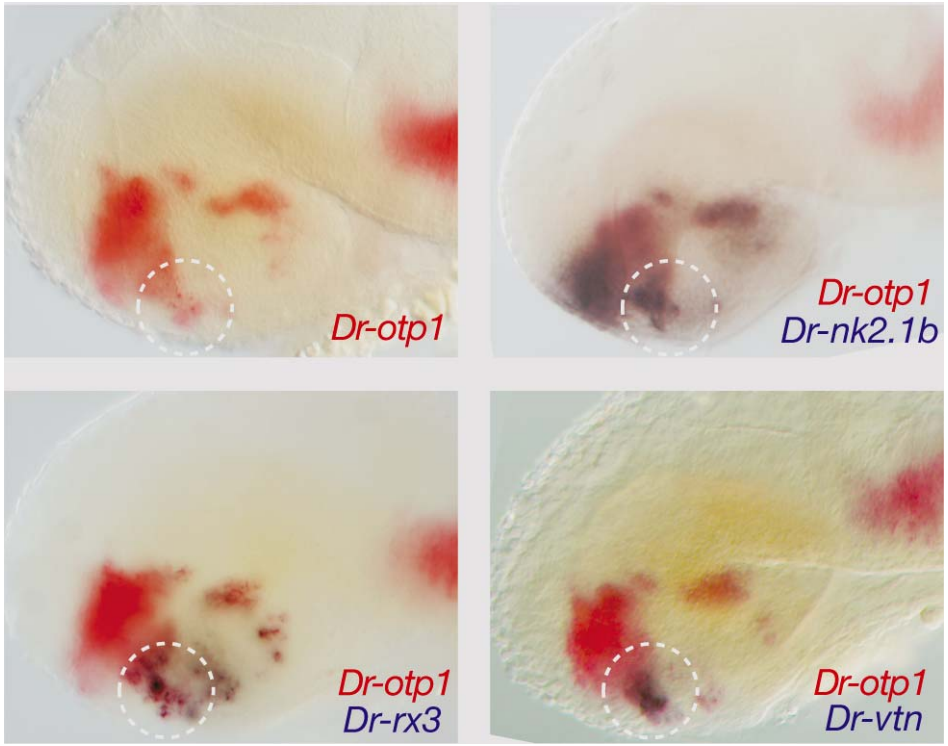


Platynereis vasotocinergic cells





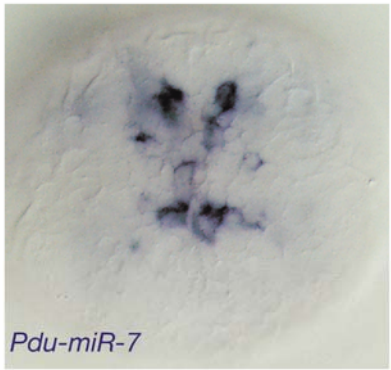
A small set of *nk2.1*+, *rx*+ and *otp*+ cells in 37hpf fish



MiR-7+ neurons populations in fish and worm forebrain include the vasotocinergic neurons

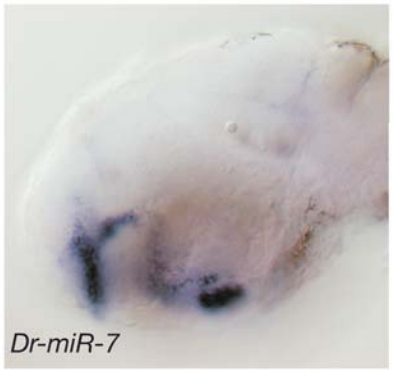
Platynereis

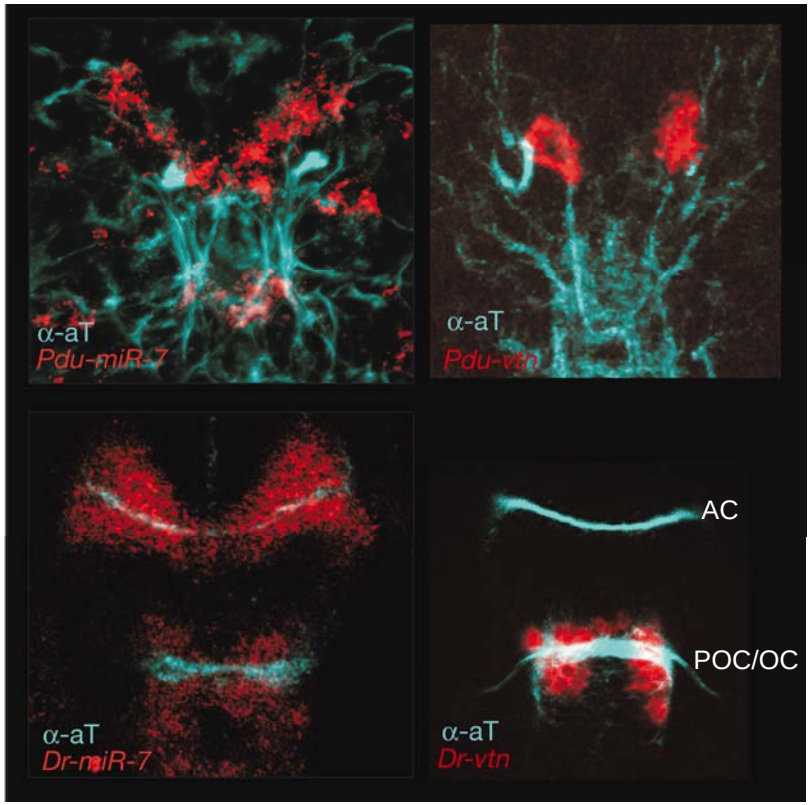
48hpf



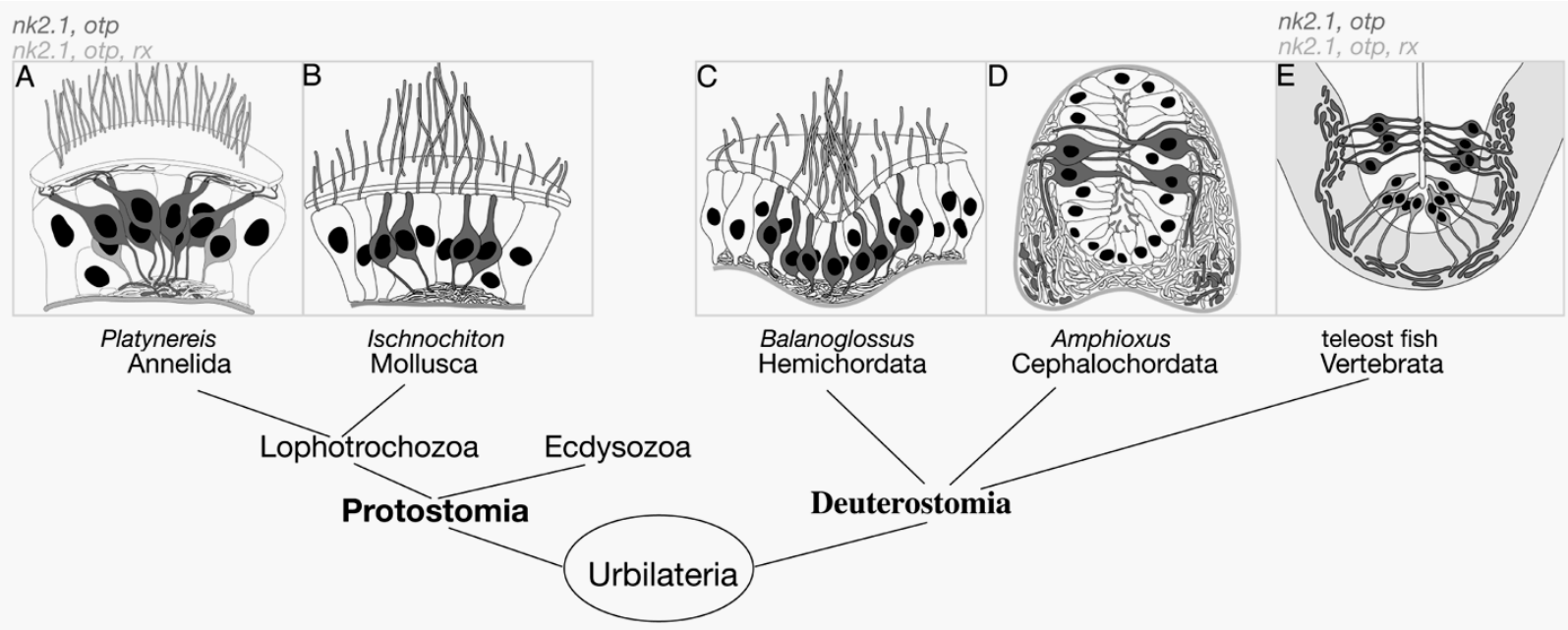
zebrafish

37hpf



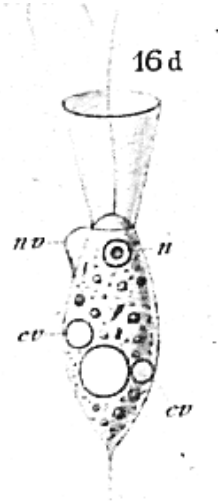


Conservation of vasotocinergic and FMRFamidergic cells

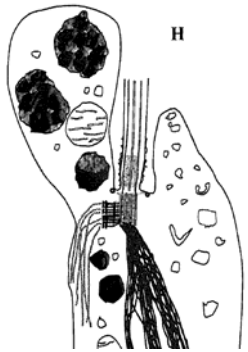


- The vasotoinergic cells are photosensitive by c-opsin expression
- seasonal and daily fluctuations of hypothalamic vasotocin content in many vertebrates

Ancient cell types are multifunctional

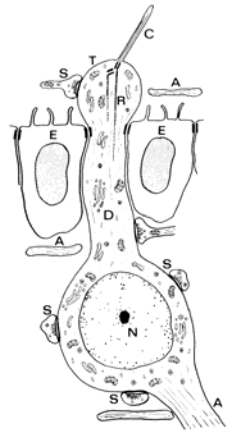


choanoflagellate



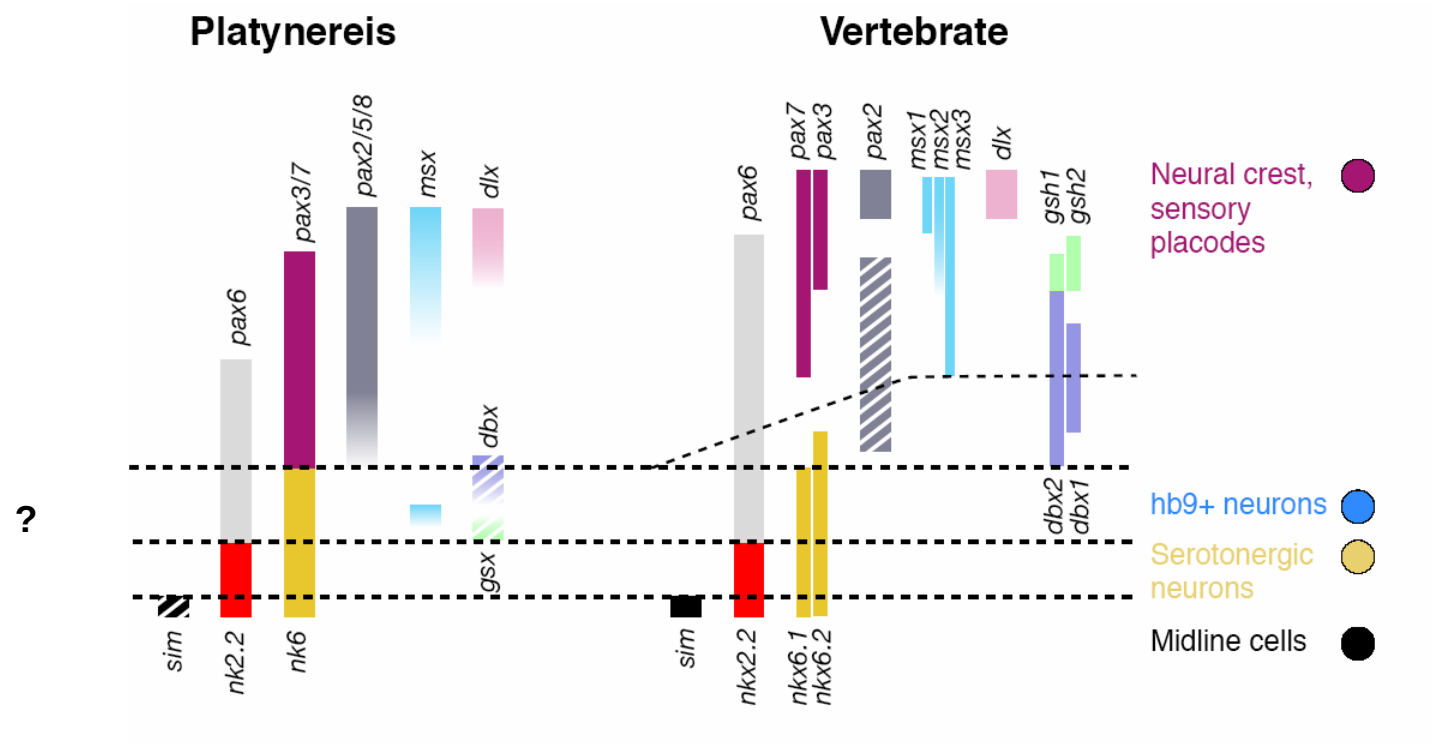
photosensitive
steering-rudder cell
(Leys and Degnan 2001)

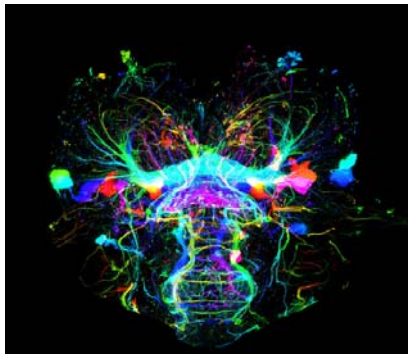
epithelial muscle cell



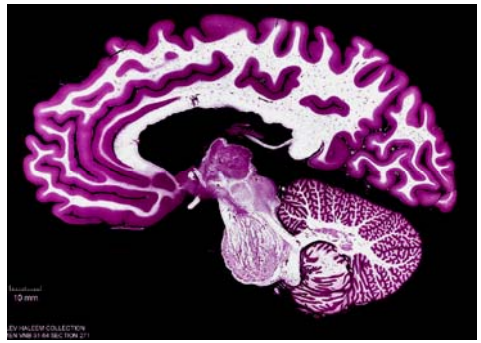
CSF-contacting neuron
(vertebrate and annelid!)

Conserved neuron types emerging from corresponding columns in polychaetes and vertebrates?





Conserved mediolateral architecture of the trunk CNS

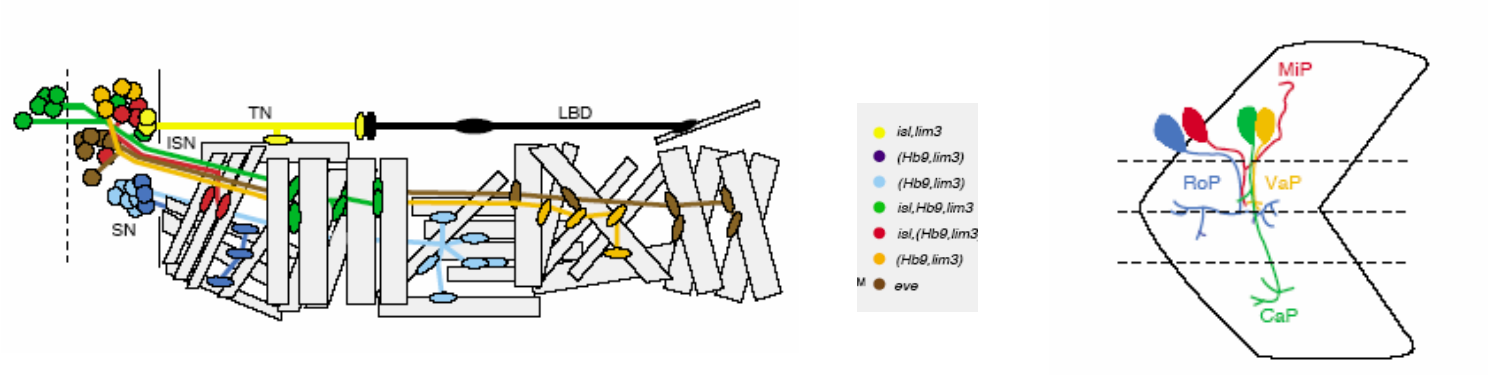


Gaspar Jekely

Cell ***129****, 277-288* (2007)

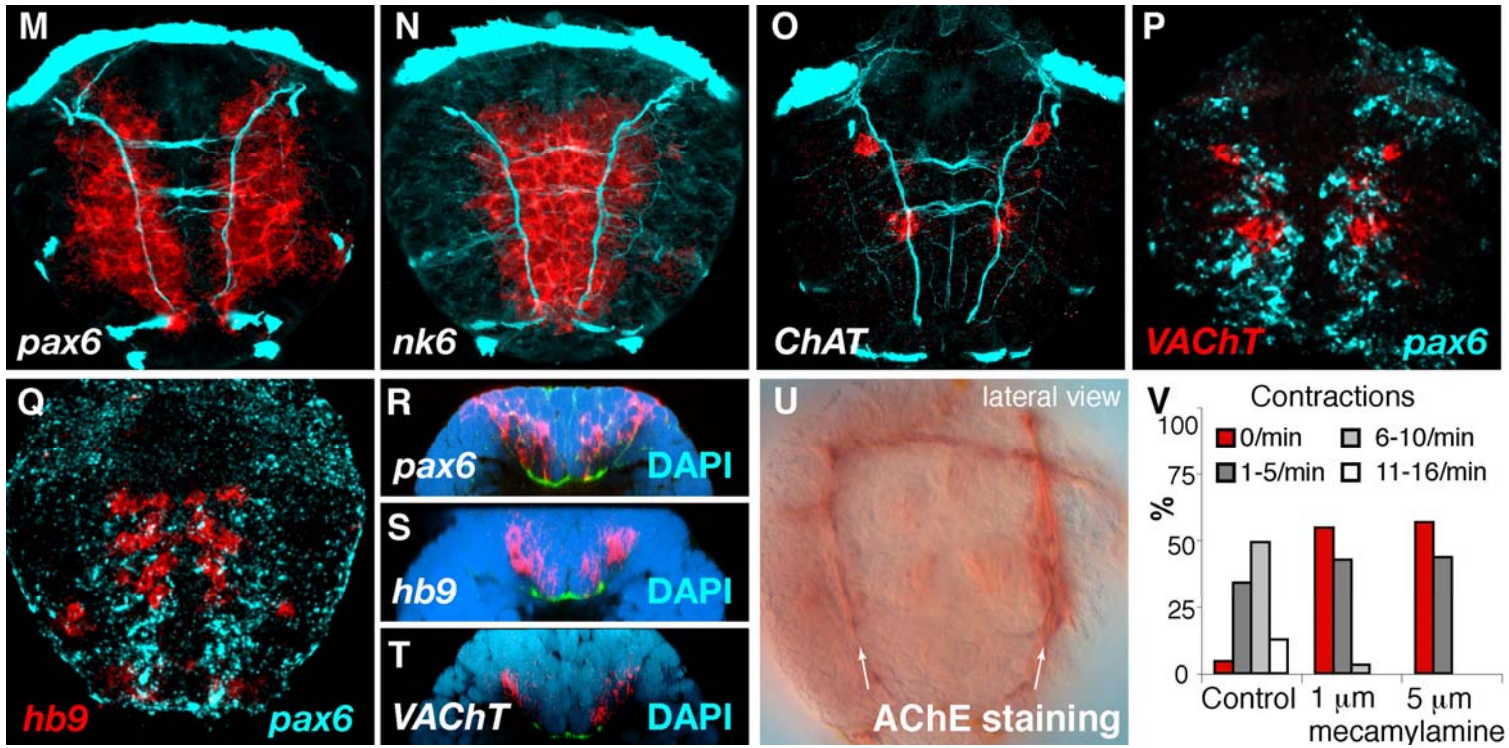
Alexandru Denes

Conservation of cell types: somatic motor neurons

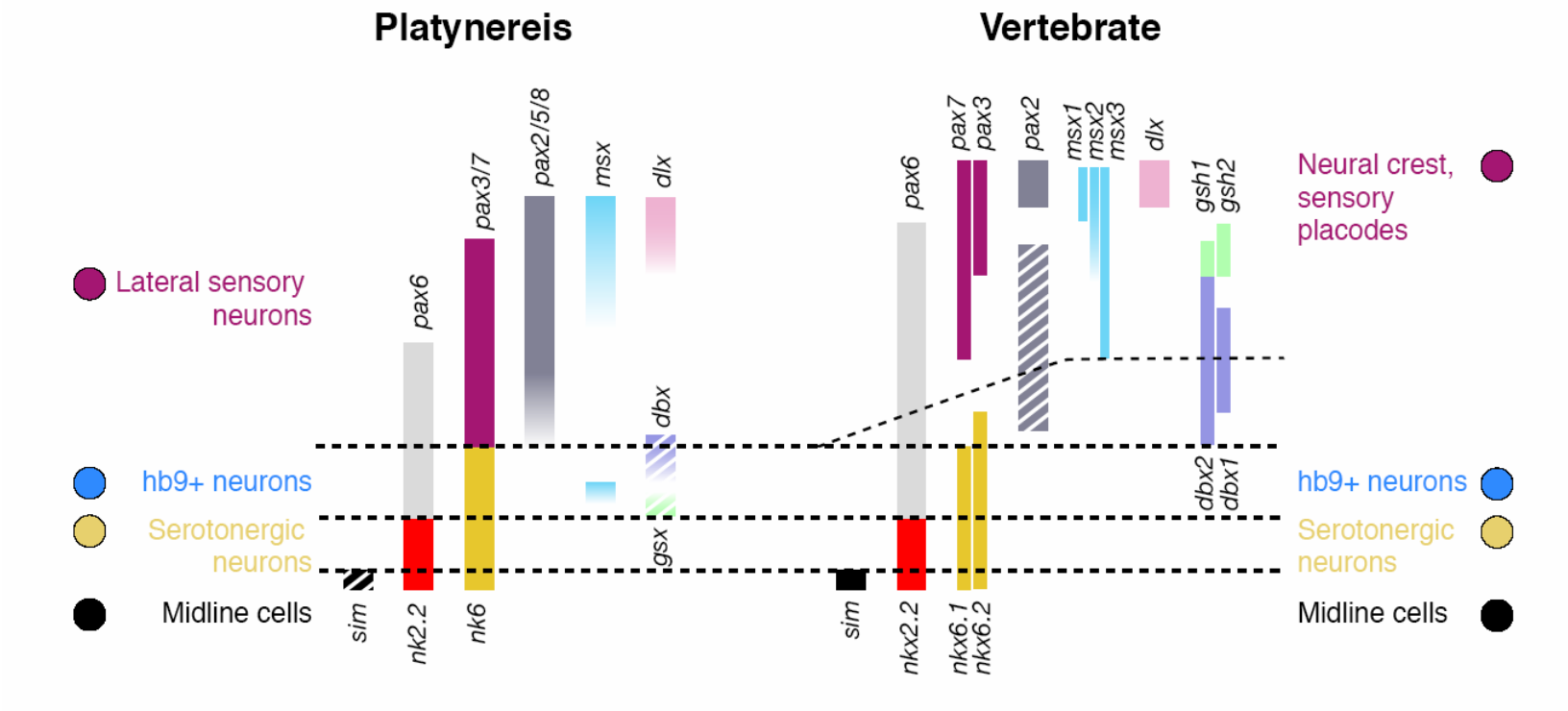


Thor and Thomas, 2002

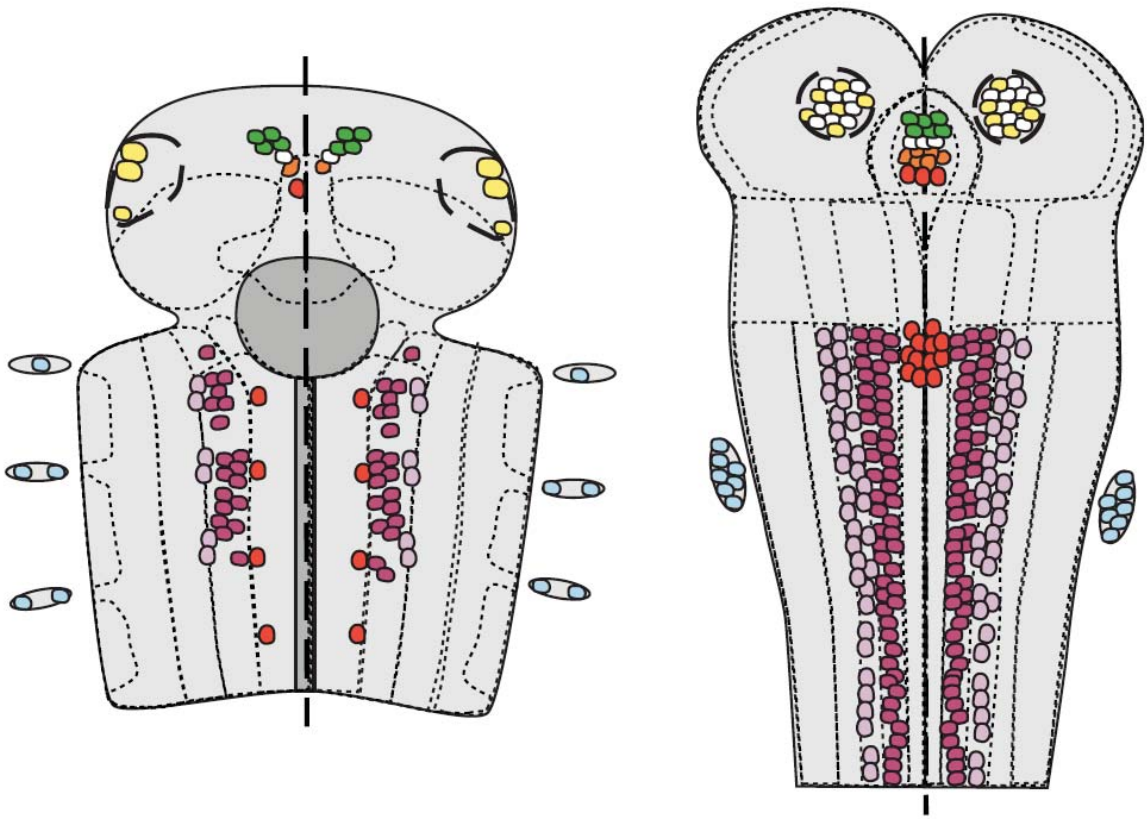
Cholinergic , *hb9*+ motoneurons emerge from the *pax6,nk6* column



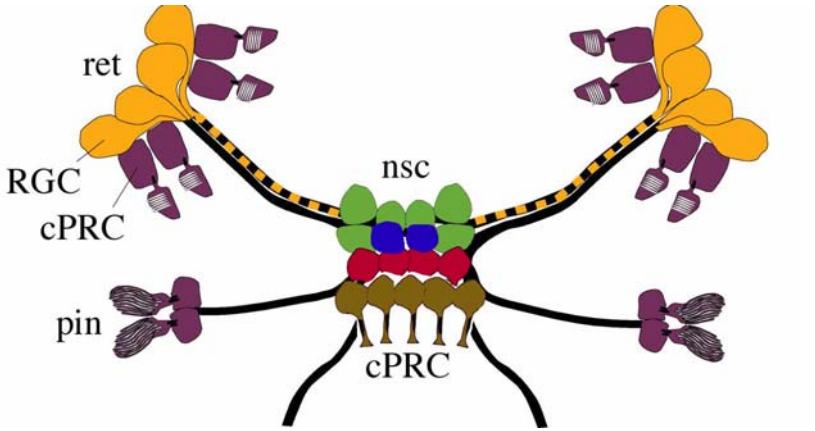
Conserved neuron types emerging from corresponding columns in polychaetes and vertebrates



The conserved molecular topography translates into a map of conserved cell types



Conservation of cell types
Comparison of vertebrate and polychaete brains

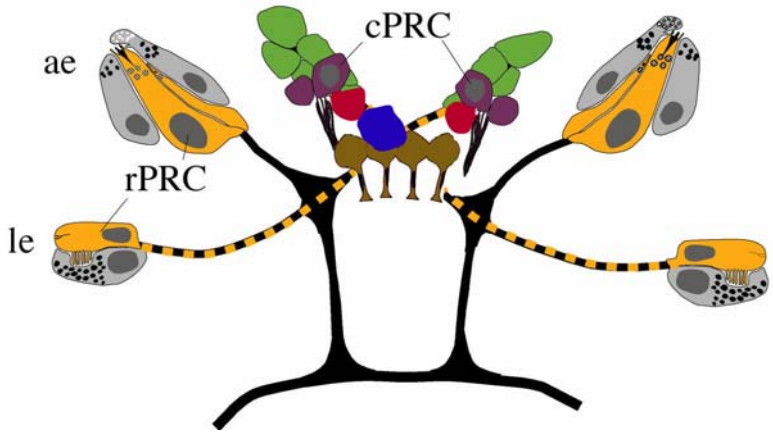


rx, nk2.1, otp
c-opsin
vasoisotocin

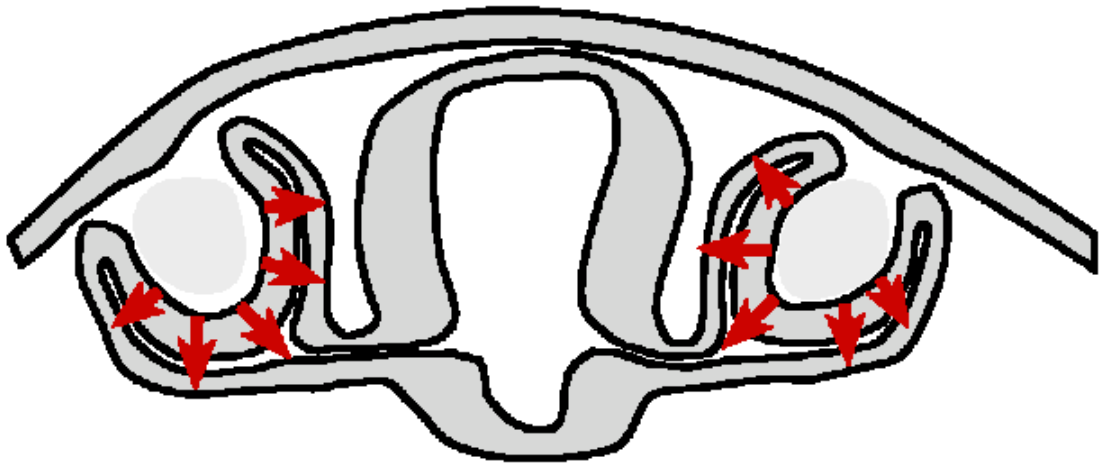
rx
c-opsin

otp
FMRF

Pax6, ath, BarH1
r-opsin



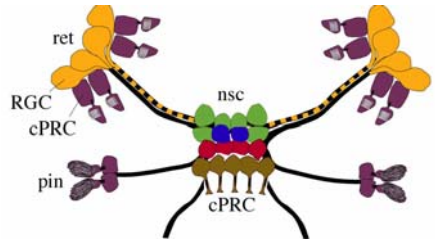
Evolution of eyes as an ‘extension’ of the forebrain



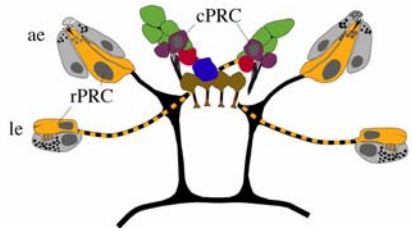
Evolution of photoreceptor cells

Rhabdomeric photoreceptors

Ciliary photoreceptors



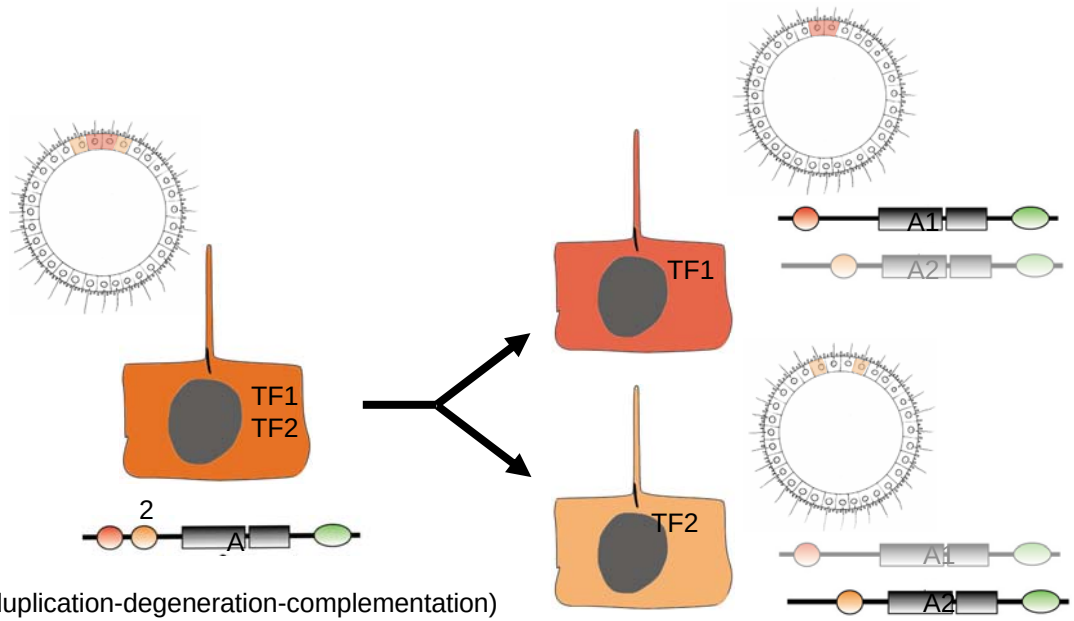
QuickTime™ and a
decompressor
are needed to see this picture.



Congruency of cell typogenetic tree and gene tree

QuickTime™ and a
decompressor
are needed to see this picture.

Gene duplication and cell type divergence



(duplication-degeneration-complementation)

Evolution of photoreceptor cells

Rhabdomeric photoreceptors

Ciliary photoreceptors

QuickTime™ and a
decompressor
are needed to see this picture.

Cell type functional divergence

Cell type functional segregation

Retinal bipolar cells and rods and cones are sister cell types

- shared regulatory signature (otx2, crx, rx)
- shared differentiation signature (recoverin, potassium channels, etc.)
- Ribbon synapses, similar G-protein cascades
- Similar cellular morphology (Landolt's clubs, retina-spanning axons,
- similar expression profile

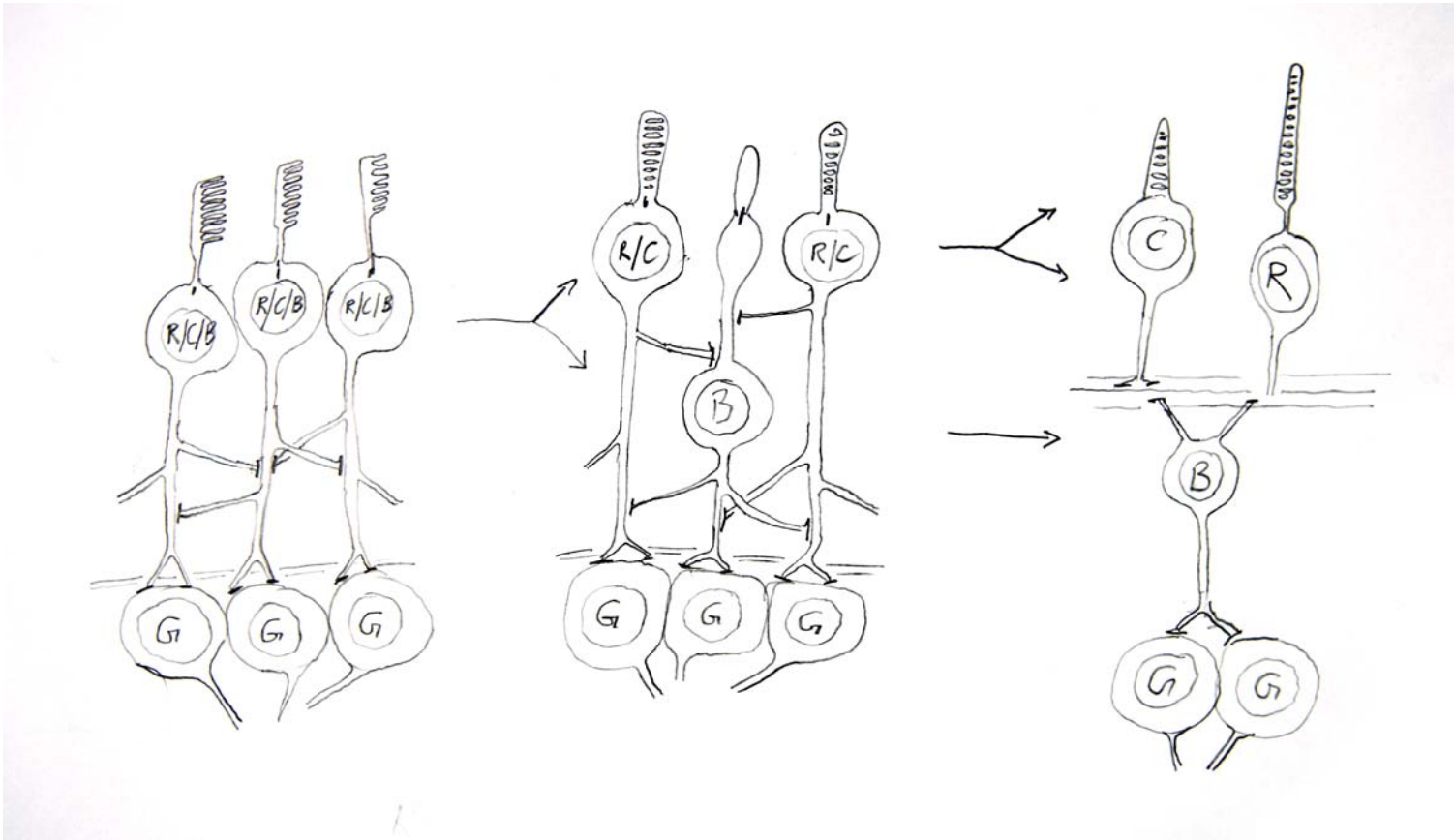
QuickTime™ and a
decompressor
are needed to see this picture.

QuickTime™ and a
decompressor
are needed to see this picture.

(Vigh et al., 2004; Lamb et al., 2008)

Blackshaw et al., 2001/2004

Evolution of neuronal circuits by neuron type segregation: rods/cones/bipolars in the vertebrate retina



Evolution of neuronal circuits by neuron type segregation nose, hypothalamus, pituitary

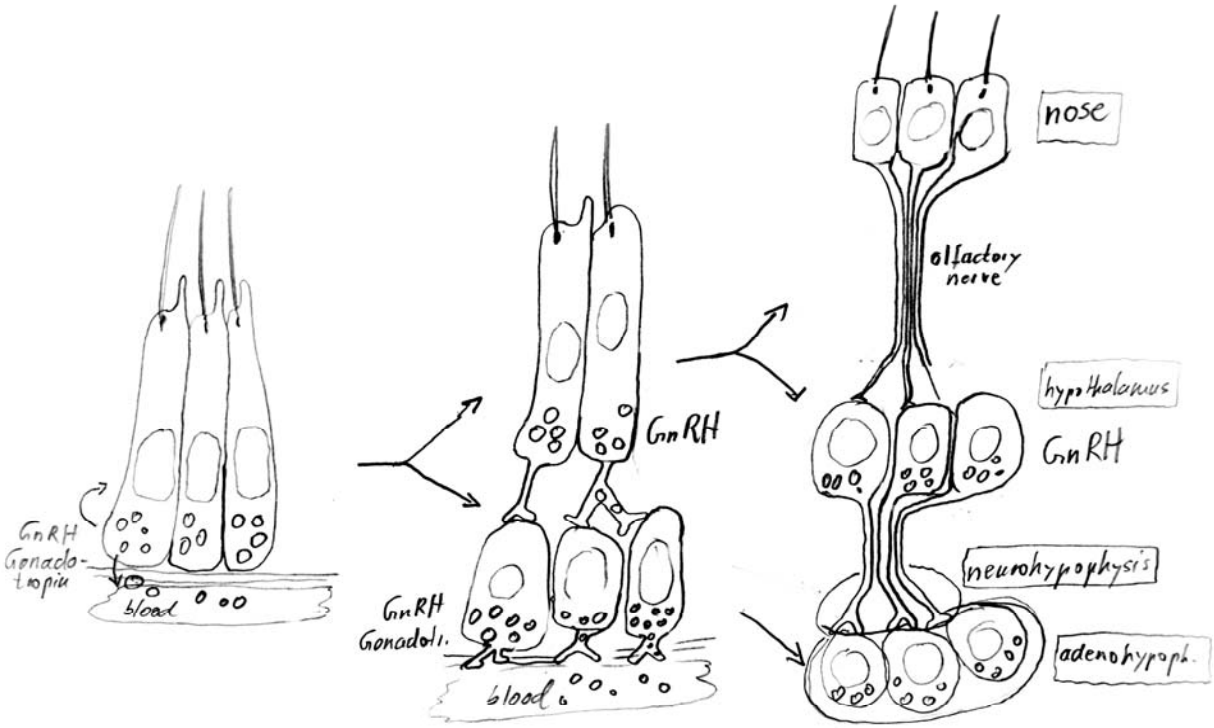
Pit1, ptx
GnRH
Gonadotropin, LH

QuickTime™ and a
decompressor
are needed to see this picture.

- Multifunctional, putative chemosensory and endocrine cells in amphioxus Hatschek's pit

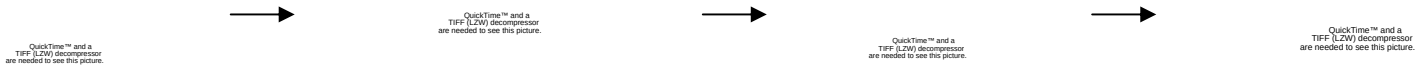
Candiani et al. 2007
Lacalli 2008

Evolution of neuronal circuits by neuron type segregation nose, hypothalamus, pituitary



- Common origin from olfacto-adenohypophyseal placode
- Migration of hypothalamal GnRH+ cells along olfactory nerve
- GnRH+ cells in olfactory system

Evolution of neuronal circuits by neuron type segregation: sensory-neuromuscular circuit

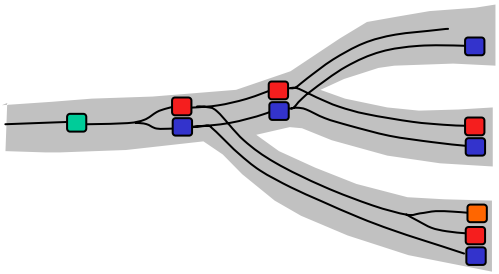


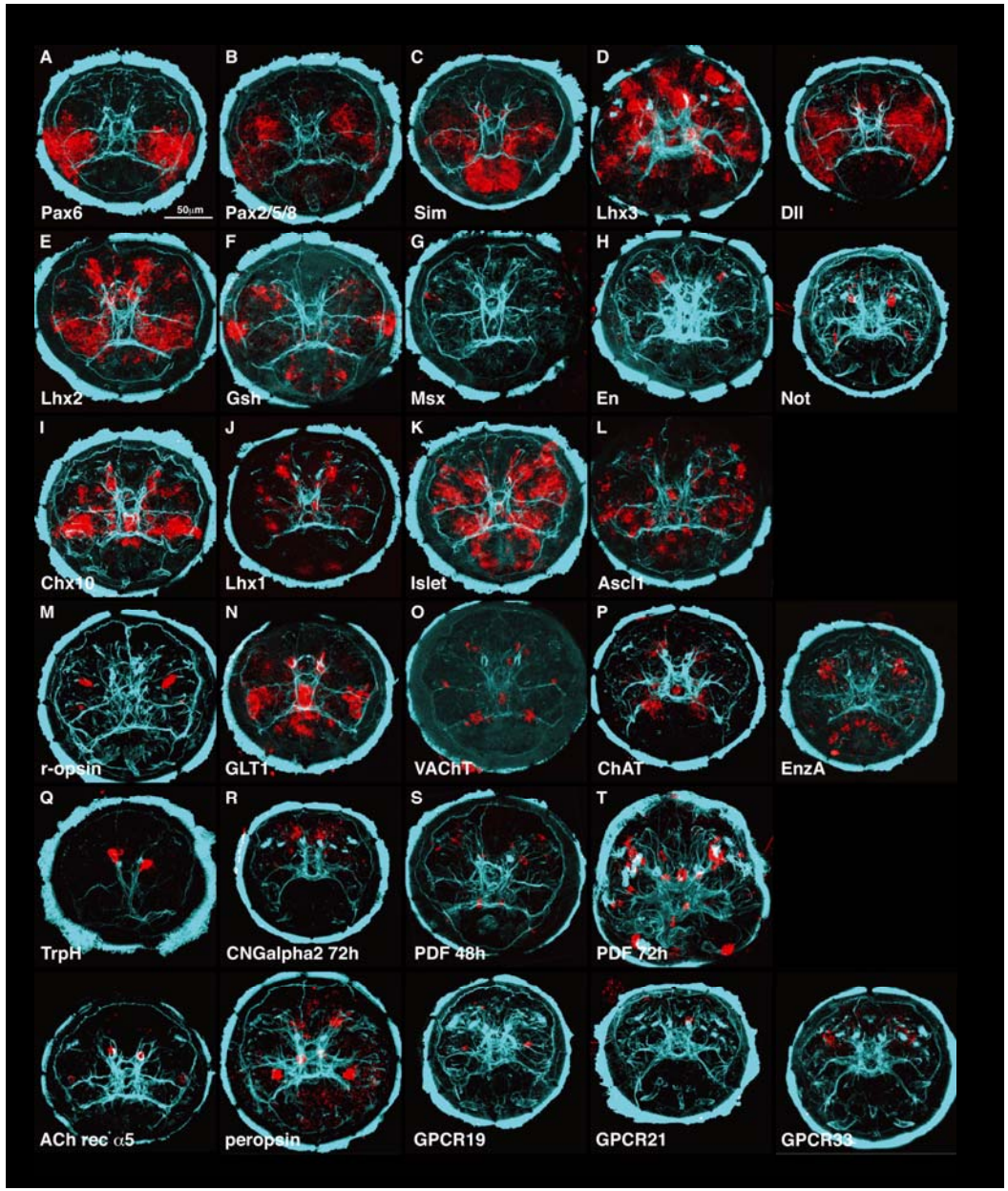
QuickTime™ and a
TIFF (LZW) decompressor
are needed to see this picture.

Perspective: complete molecular fingerprints at large scale

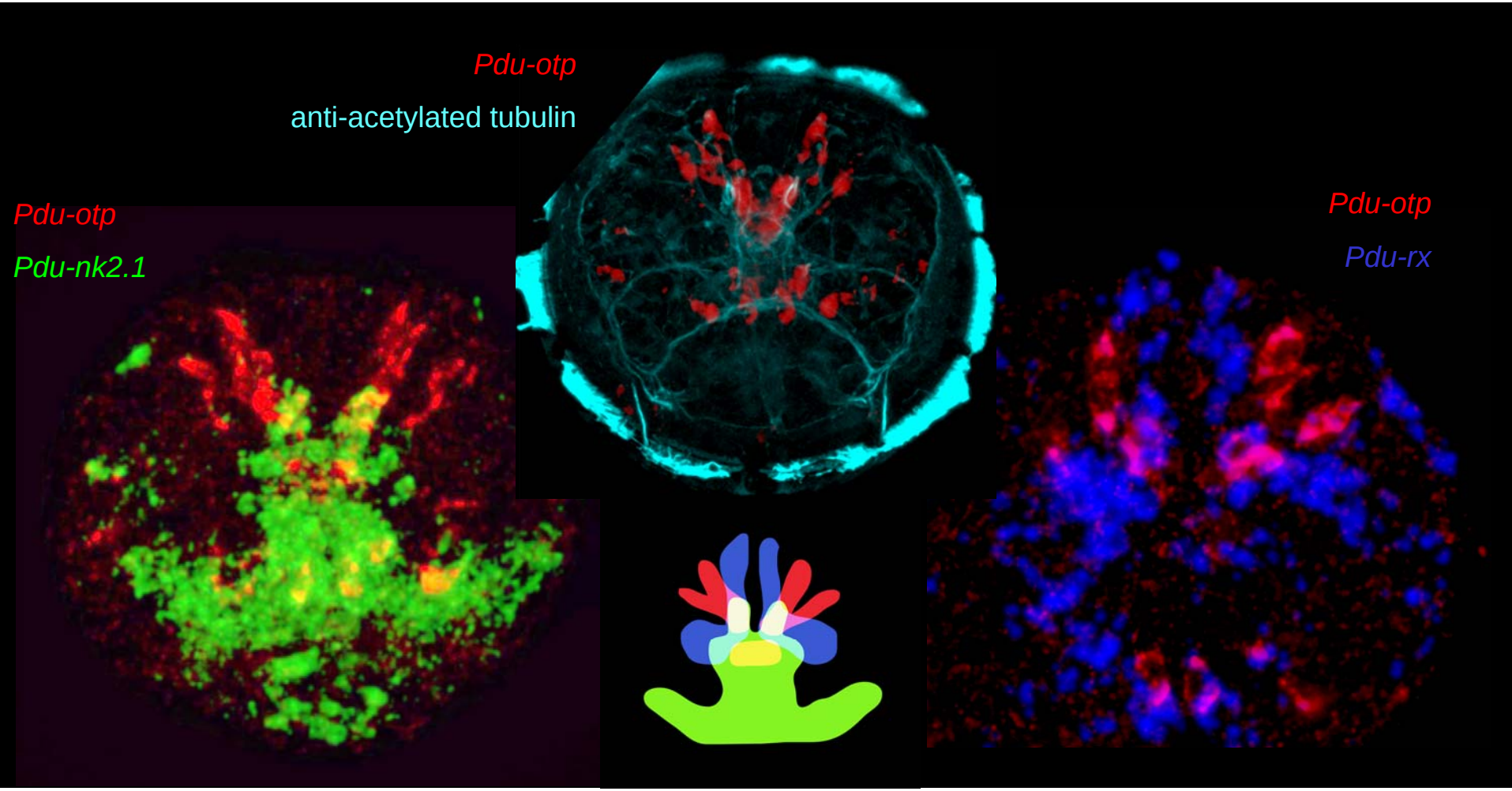
How can we get complete?

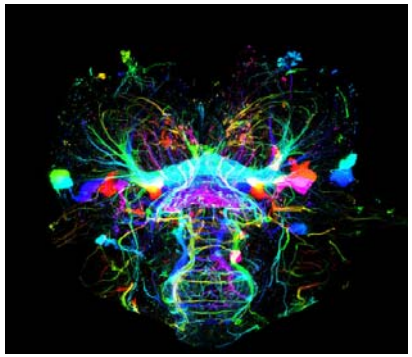
Complete single cell expression profiling in as many species as possible



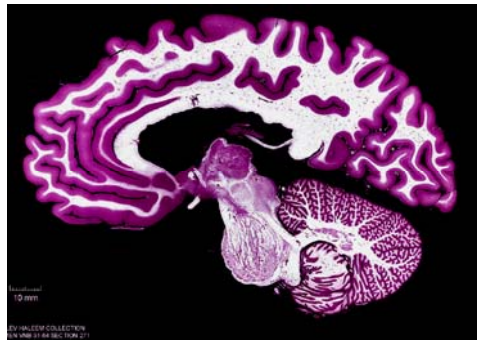


If this is a whole postdoctoral work, how can we speed up?



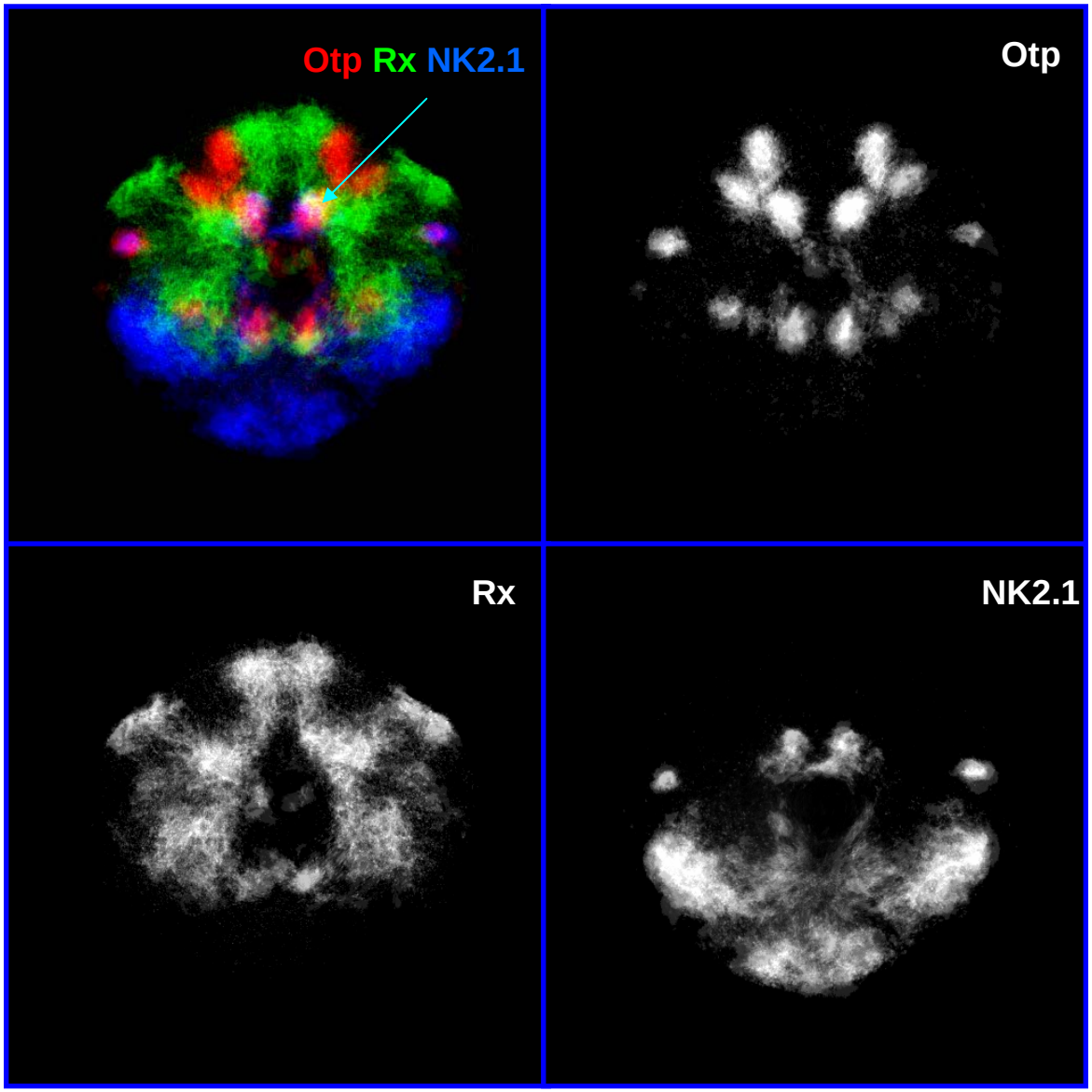


Single cell expression profiling



Raju Tomer

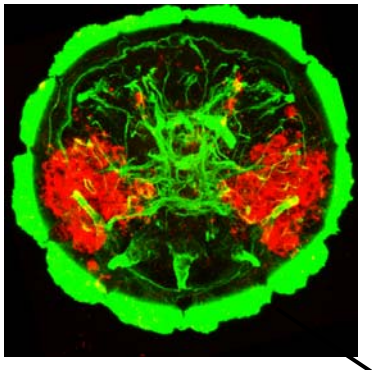
Wholemount In Silico Expression Profiling

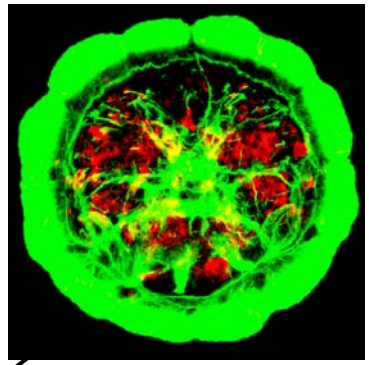


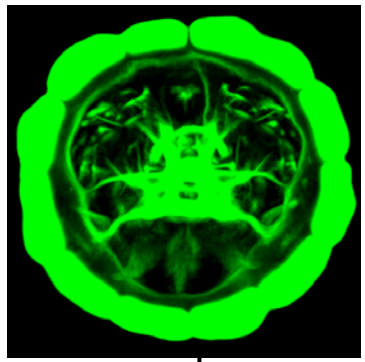
The developing axonal scaffold is highly stereotypic

QuickTime™ and a
Video decompressor
are needed to see this picture.

Image registration







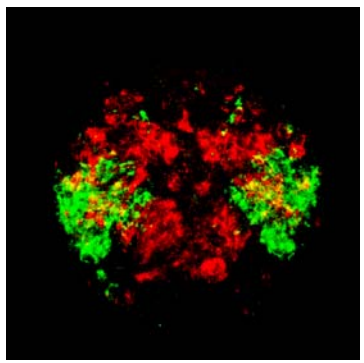
Affine (Rigid)

Warp (Non-Rigid)

Average Brain Model

Normalized Mutual Information

$$Y(F,G) = \frac{\sum_{f_a \in F(\mathbf{X}_0)} P(f_a)\log(P(f_a)) + \sum_{g_b \in G(\mathbf{X}_0)} P(g_b)\log(P(g_b))}{\sum_{(f_a,g_b)\in F(\mathbf{X}_0)\times G(\mathbf{X}_0)} P(f_a,g_b)\log(P(f_a,g_b))}$$



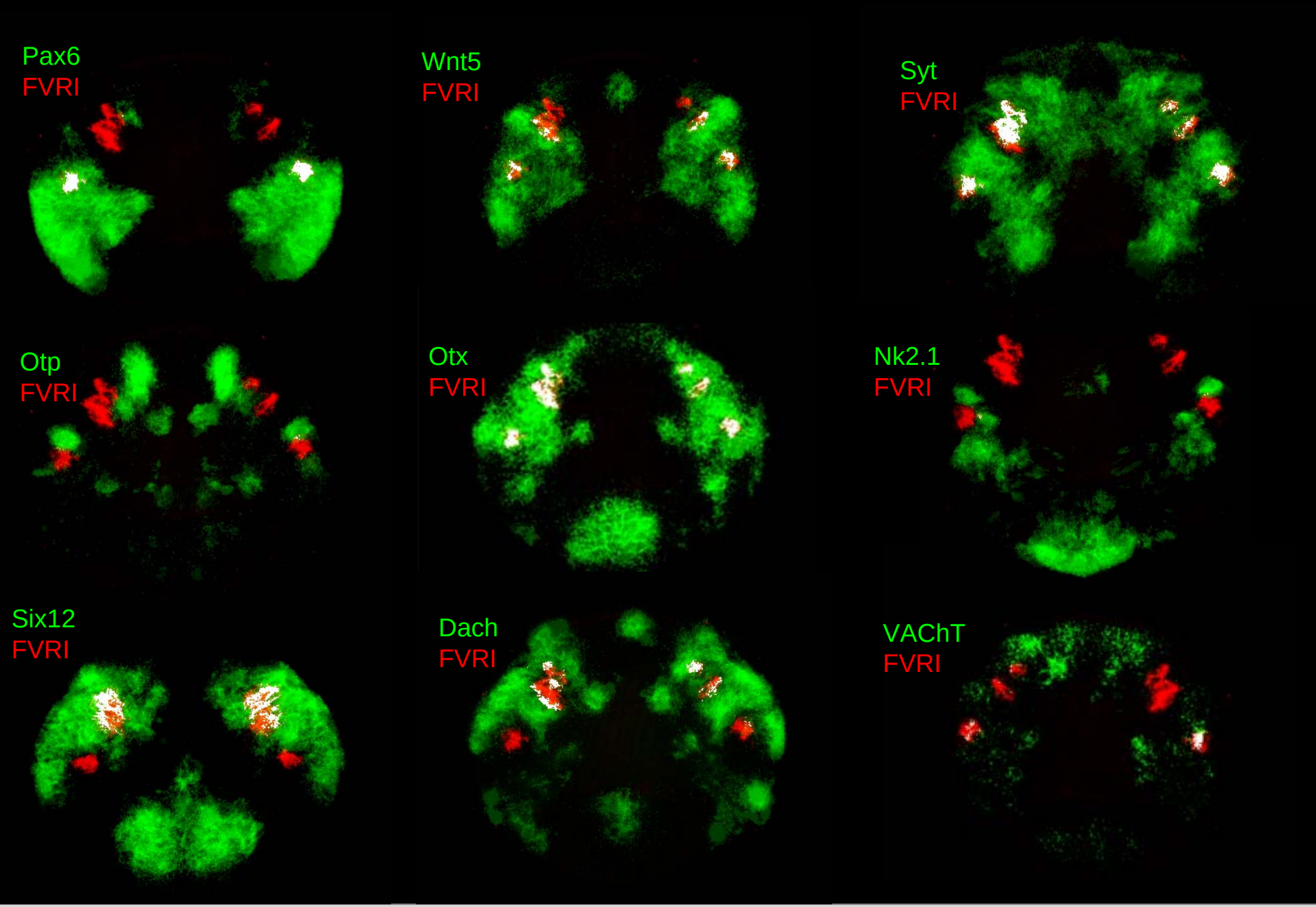
Holden et. al. 2000

Rohlfing 2005

Jefferis et. al. Cell 2007

Matching expression patters with high precision:
FVRI @ 48hpf; acetylated tubulin alignment

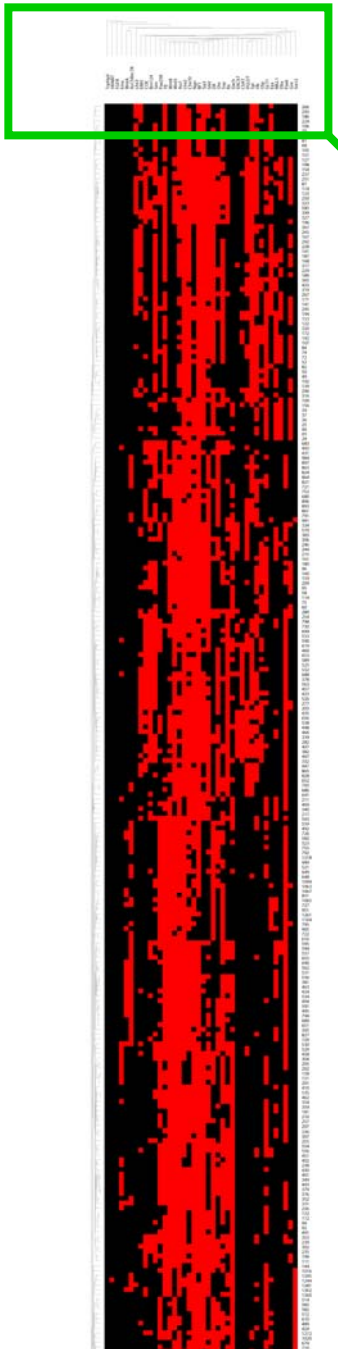
QuickTime™ and a
Cinepak decompressor
are needed to see this picture.

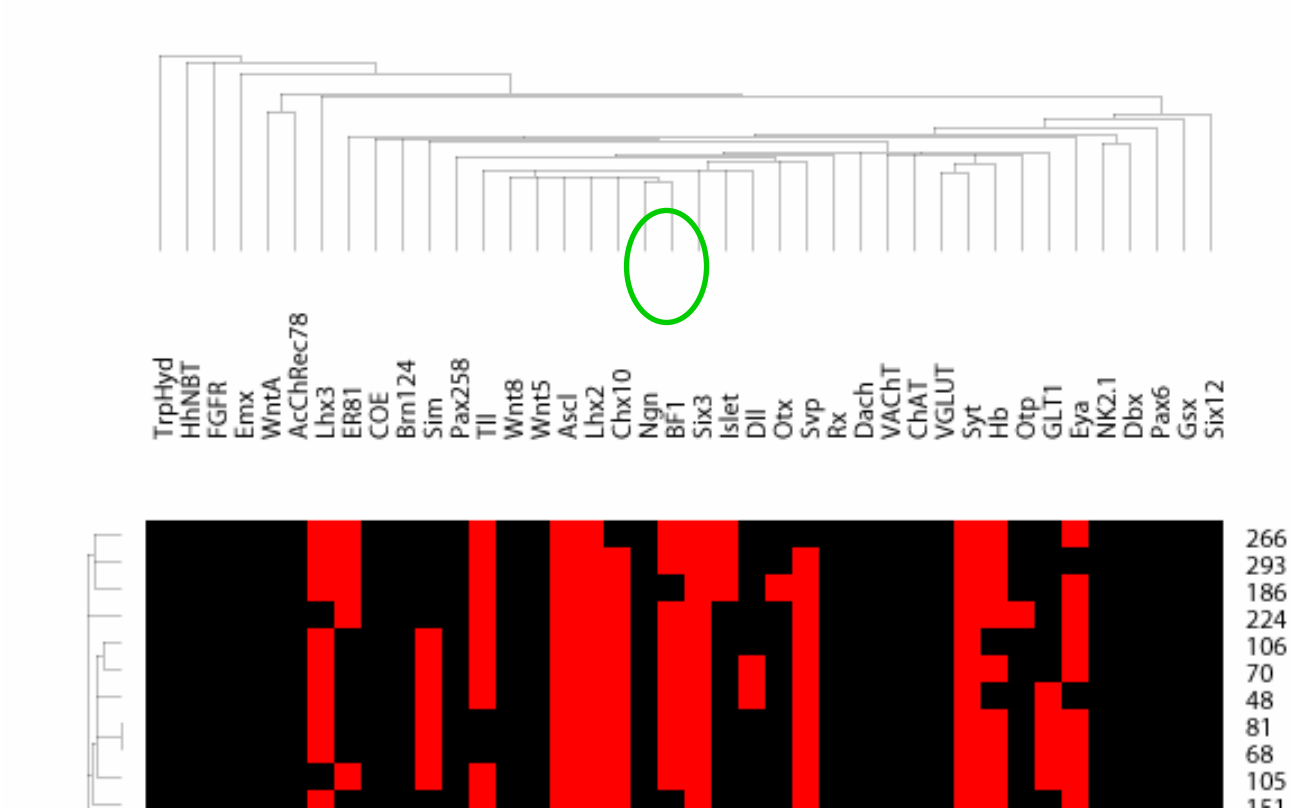

Pax6
FVRI
Wnt5
FVRI
Syt
FVRI
Otp
FVRI
Otx
FVRI
Nk2.1
FVRI
Six12
FVRI
Dach
FVRI
VAChT
FVRI

Cellular model of the *Platynereis* brain

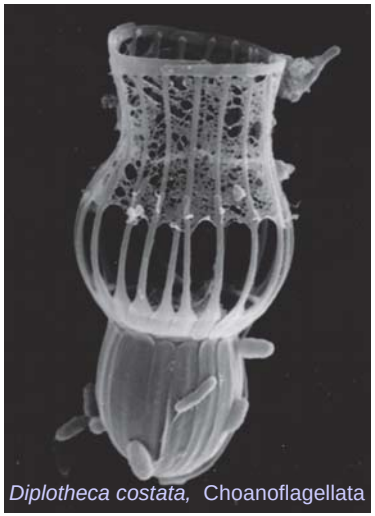
QuickTime™ and a
Video decompressor
are needed to see this picture.

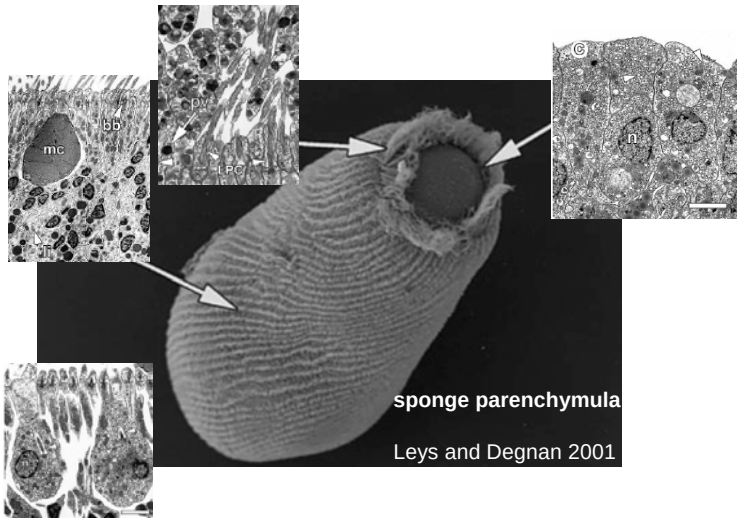
Single cell expression profiling

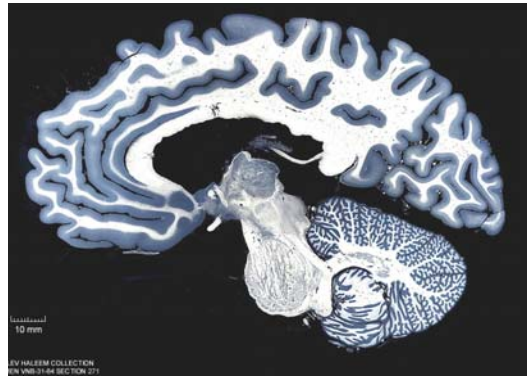


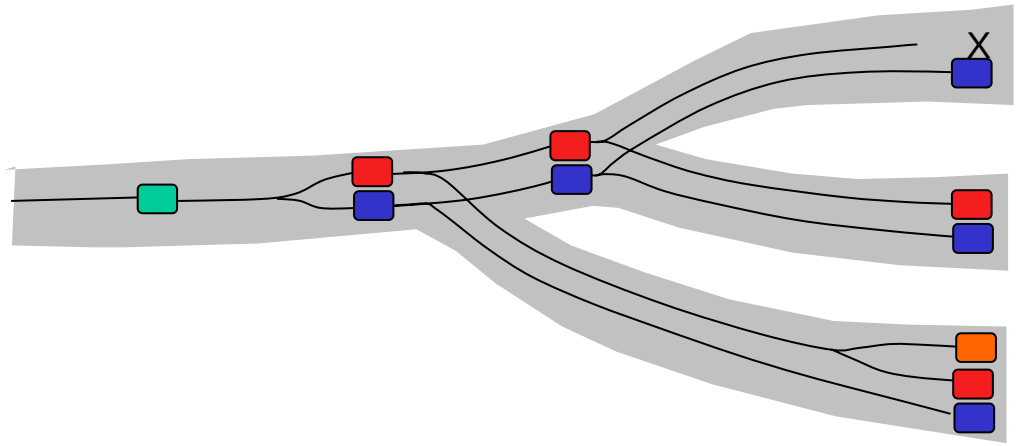


Cell type diversification


Diplotheca costata, Choanoflagellata

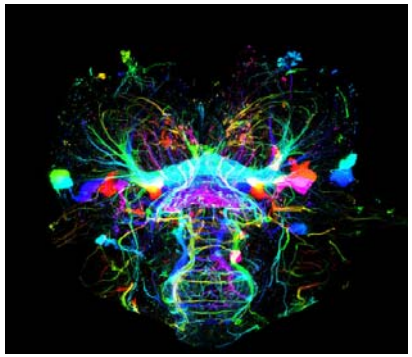




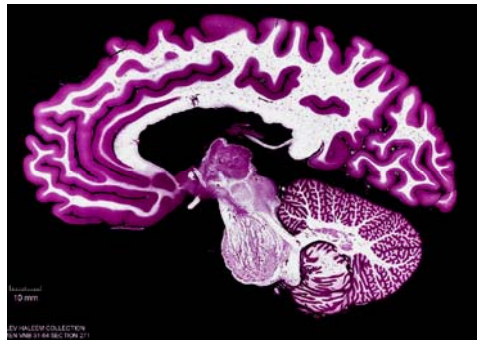


homologous cell types

sister cell types



Platynereis phototaxis



QuickTime™ and a
TIFF (Uncompressed) decompressor
are needed to see this picture.

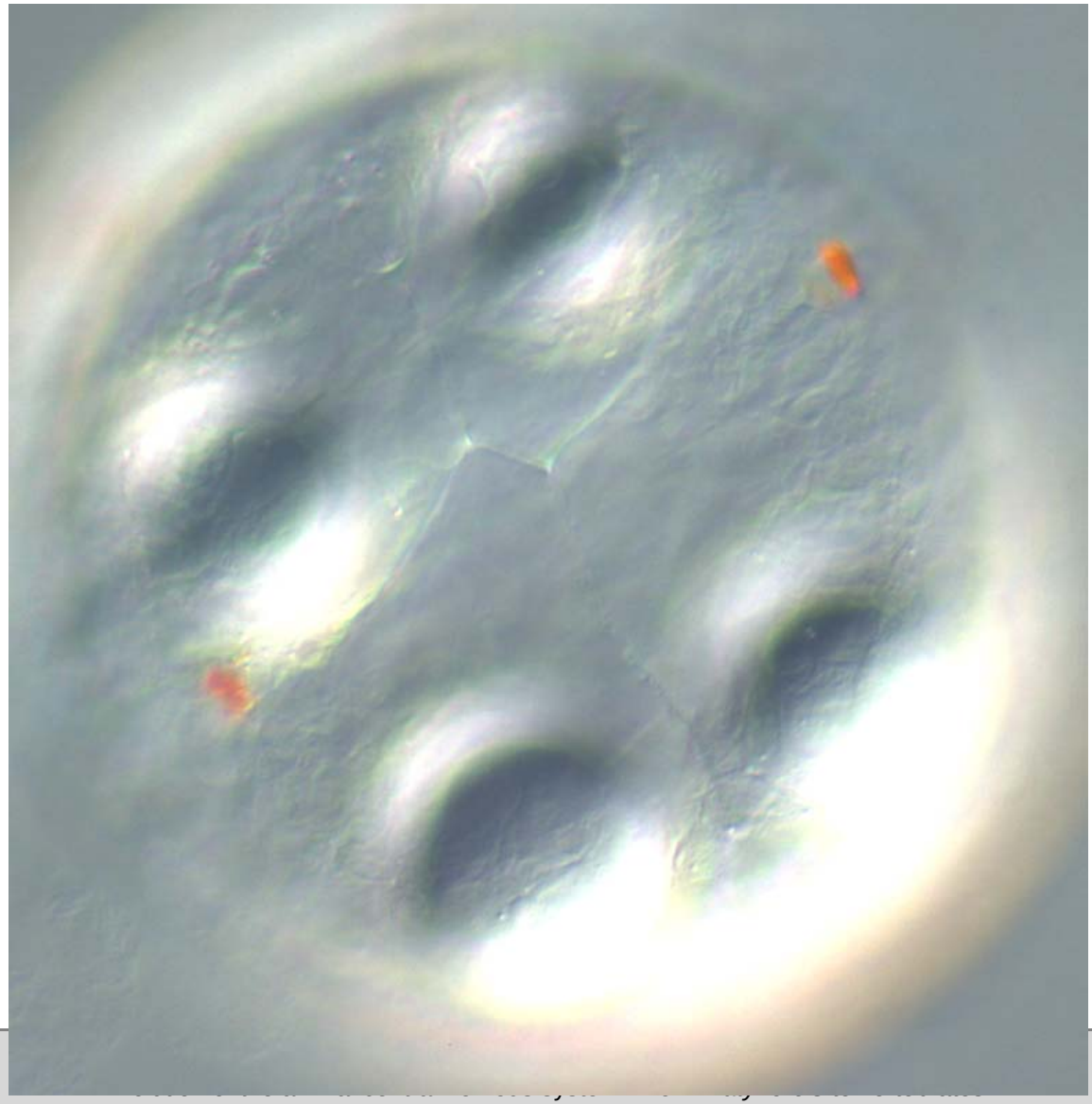
collaboration
François Nédélec

Gaspar Jekely

QuickTime™ and a
TIFF (Uncompressed) decompressor
are needed to see this picture.

collaboration
Julien Colombelli,
Stelzer lab

Players: the larval eyes…



…and the ciliary band - the larval motor

QuickTime™ and a
Cinepak decompressor
are needed to see this picture.

ImagingSource

DMK 21BF04
Digital camera

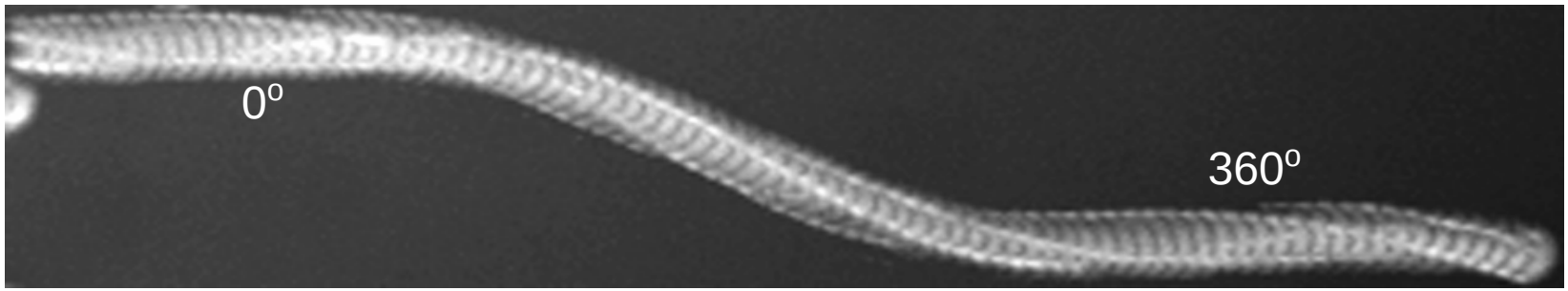
up to 60 frames per second

Platynereis phototaxis

QuickTime™ and a
Animation decompressor
are needed to see this picture.

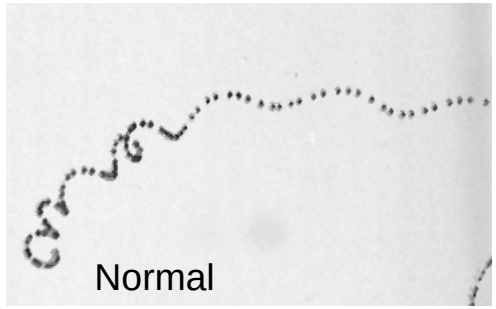
QuickTime™ and a
Animation decompressor
are needed to see this picture.

Helical swimming



Are the larval eyes really responsible?

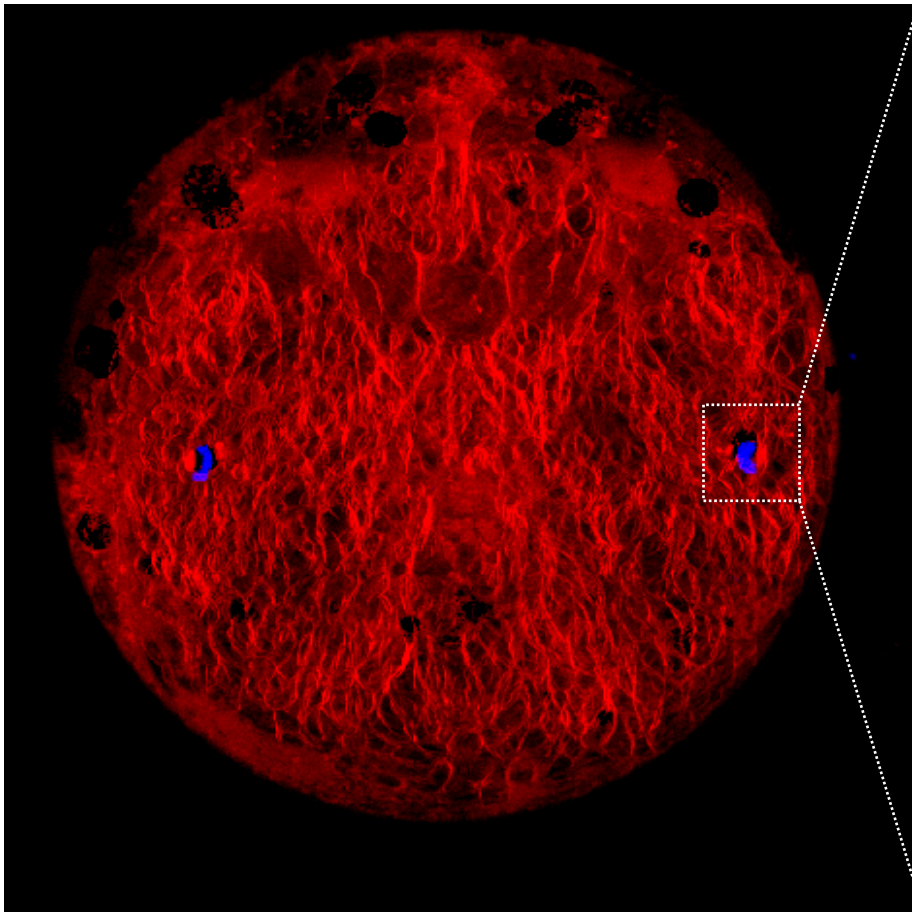
Laser ablation of larval eyes

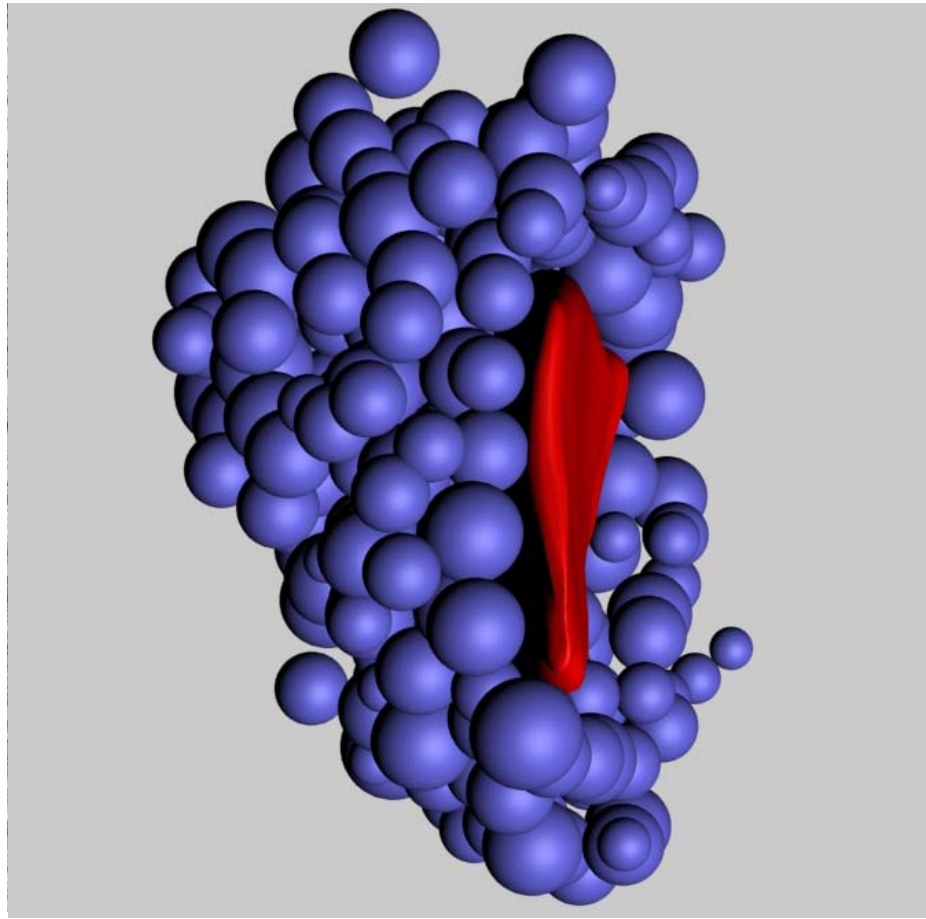


QuickTime™ and a
Sorenson Video decompressor
are needed to see this picture.

Both eyes ablated

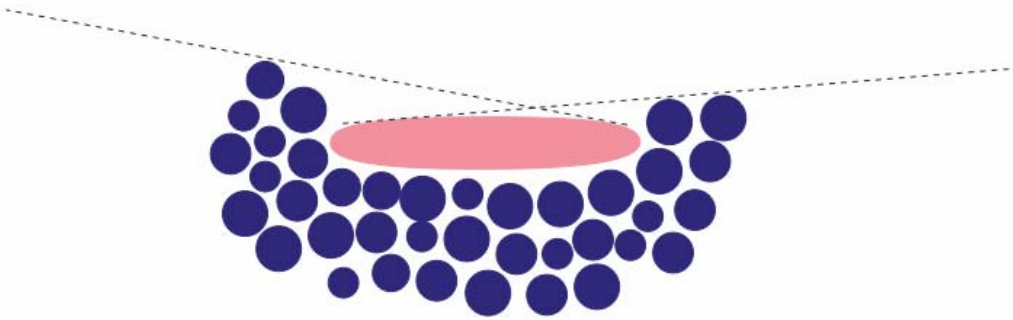
Structural constraints - simplest direction sensing eyes



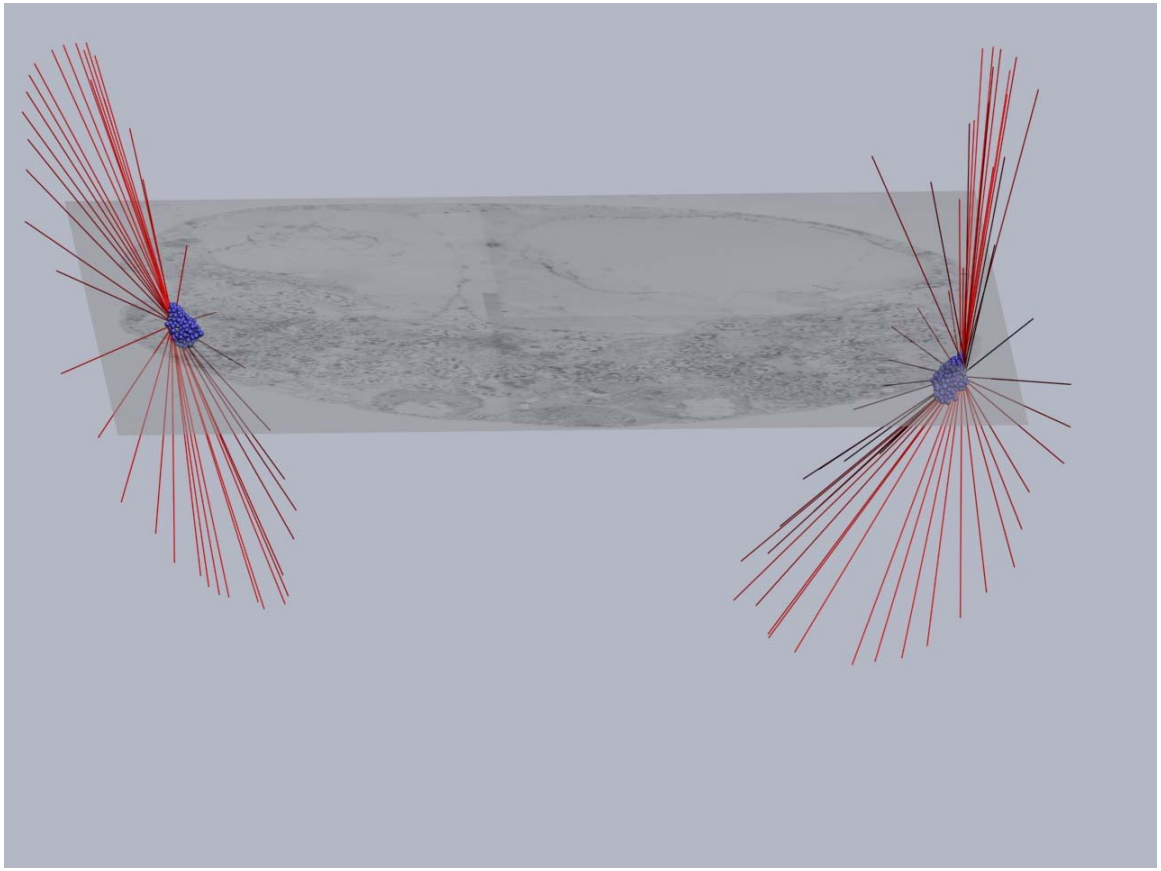


Bodipy propionic acid **Eye pigment**

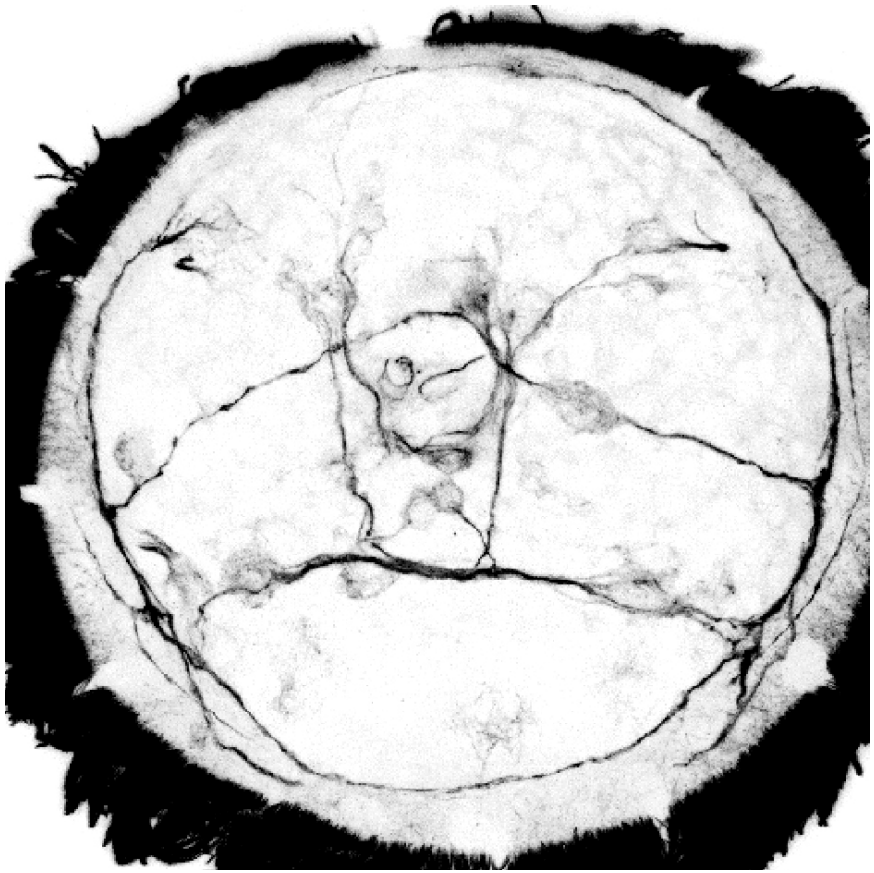
Determine viewing angle

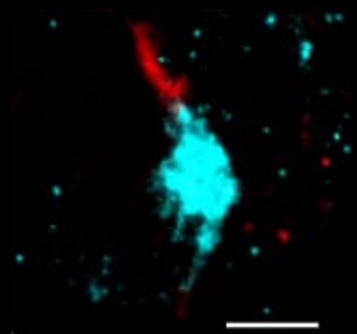


Integrate into larval morphology

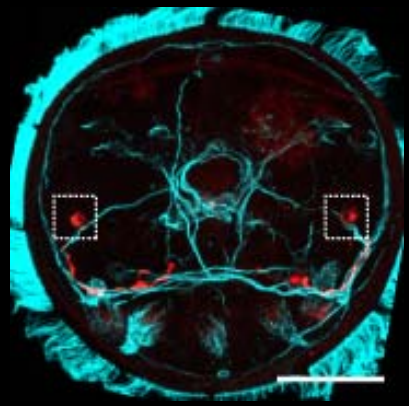


Larval eyes innervate the prototroch

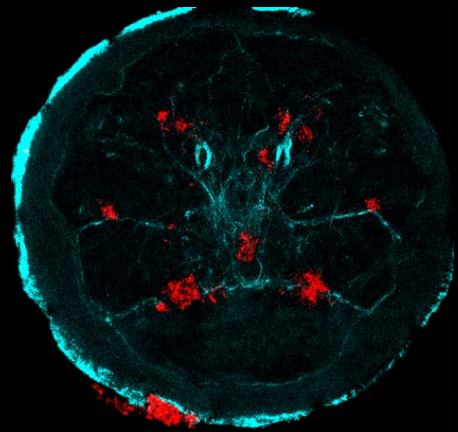




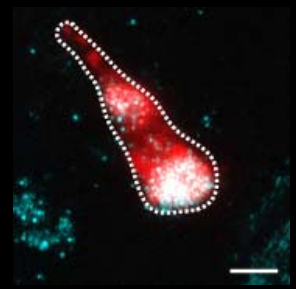
α-FVRIamide
rho-phalloidin



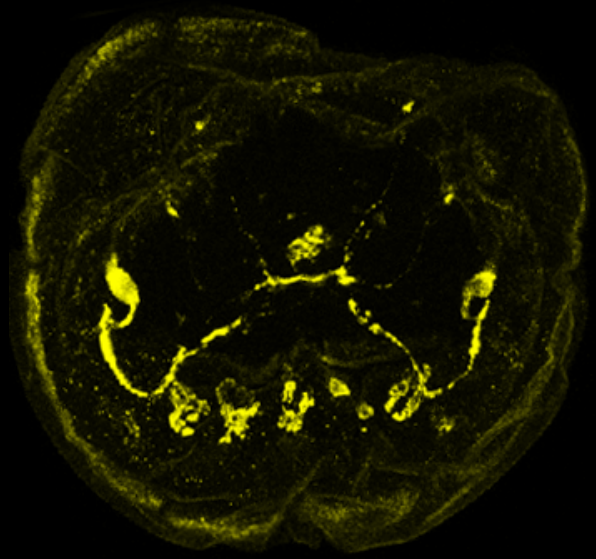
α-FVRIamide
α-acTub



VAChT
α-acTub

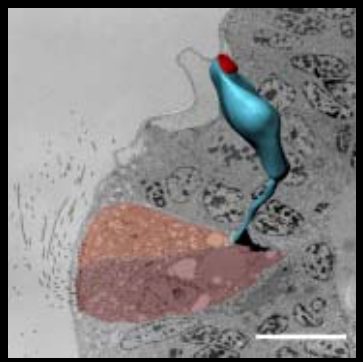


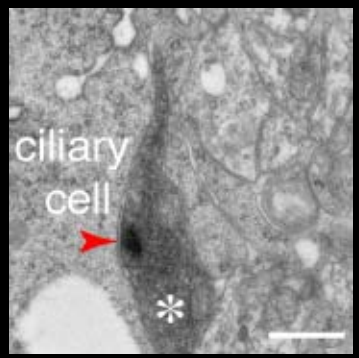
VAChT
FVRI

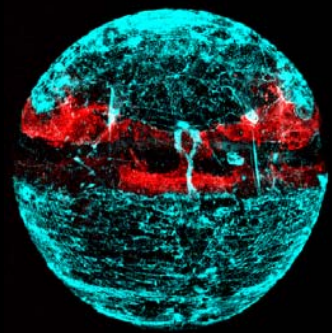


Cholinergic motoneuron marker

AChRec-α9/10 α-acTub

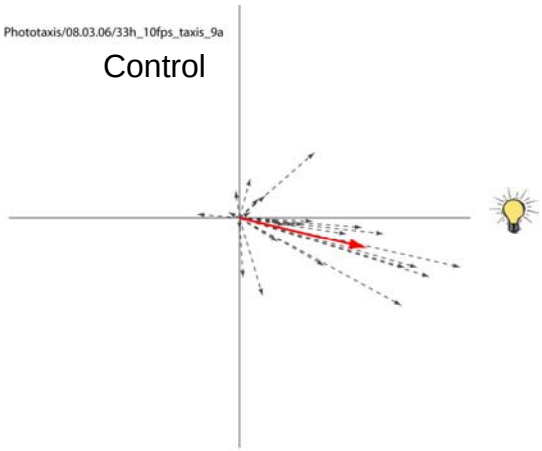




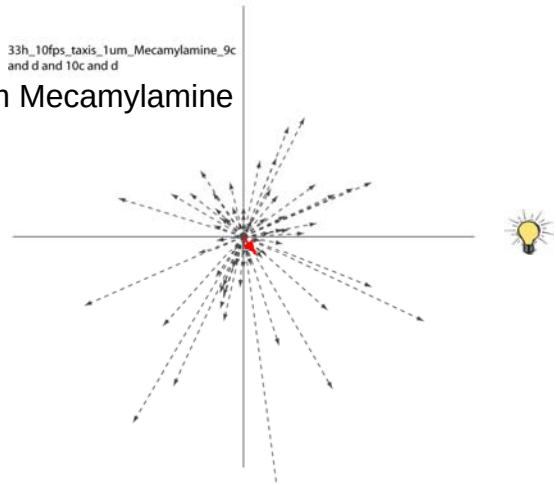


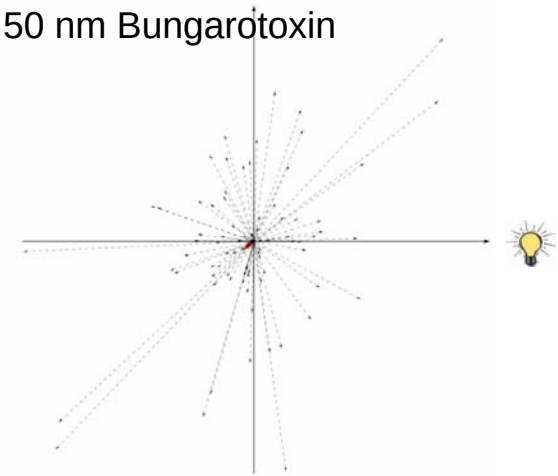
FITC-bungarotoxin
rho-phalloidin

Acetylcholine regulates phototactic steering



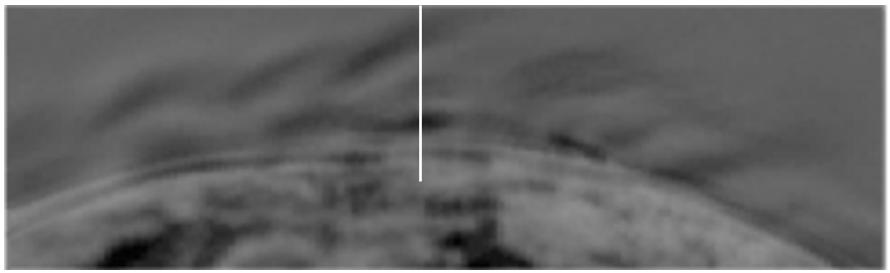
QuickTime™ and a
Animation decompressor
are needed to see this picture.

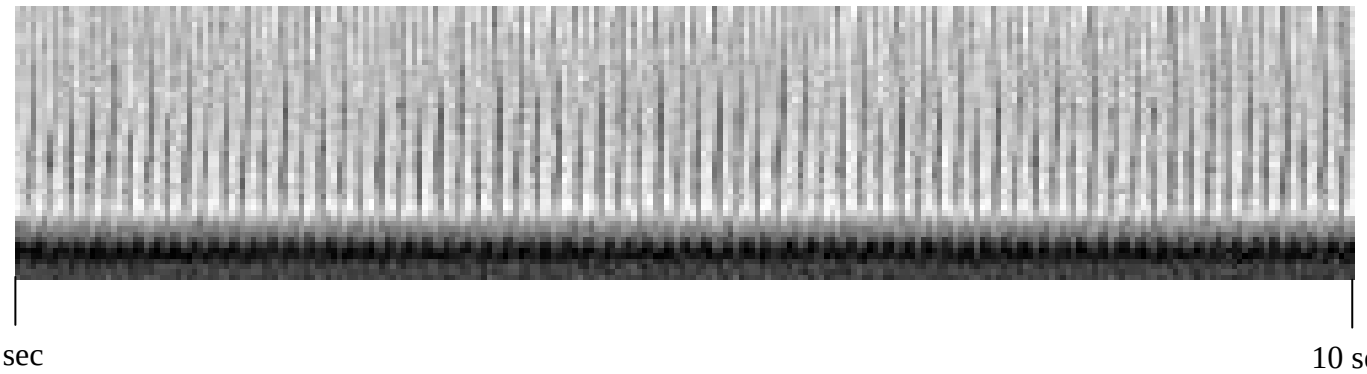




How does phototactic steering work?

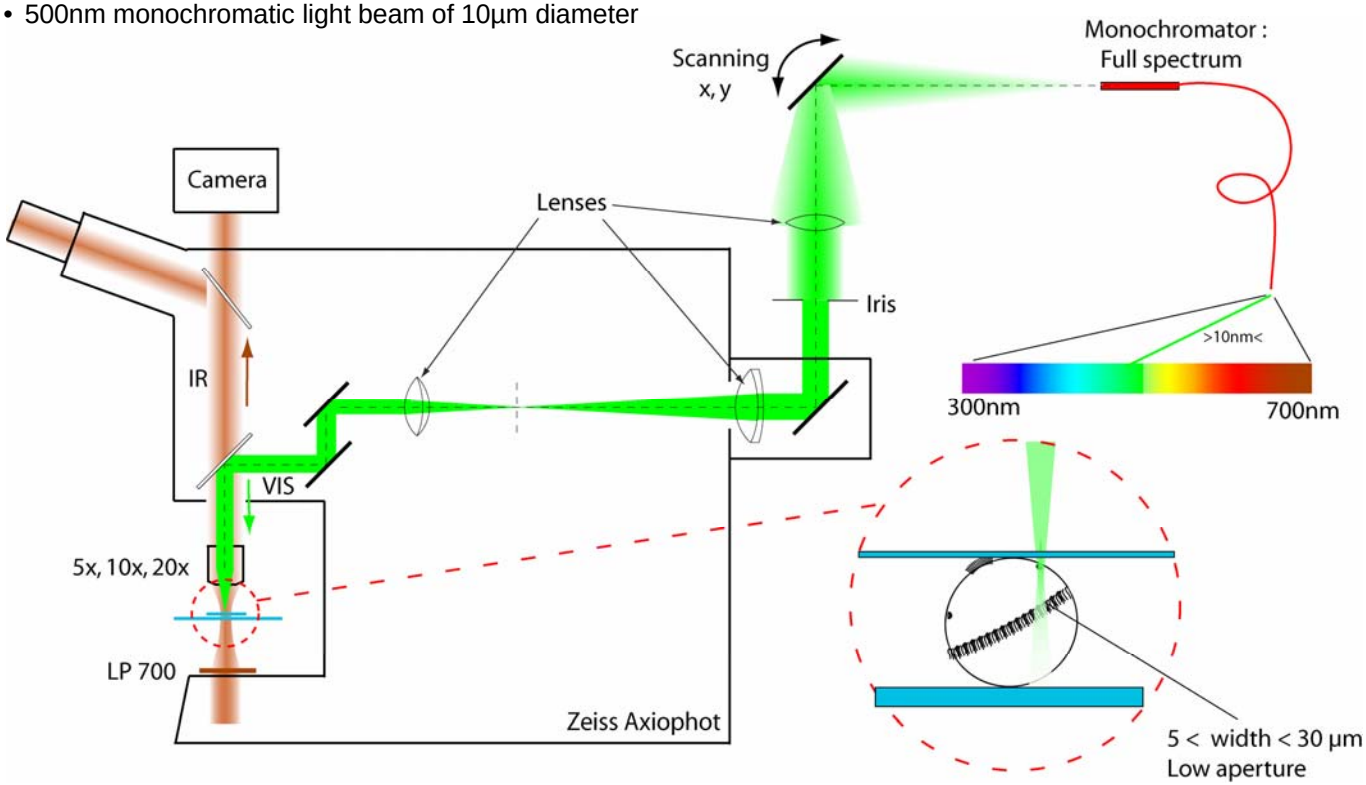
Analysis of ciliary beating





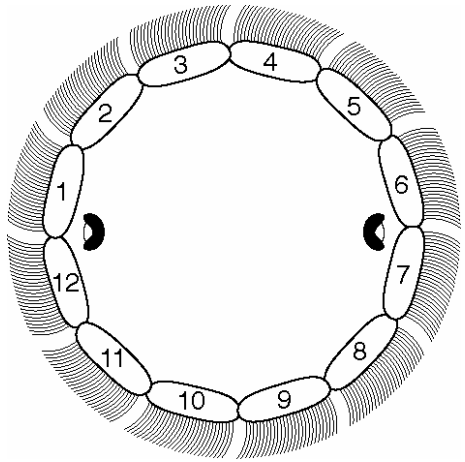
Selective eye illumination

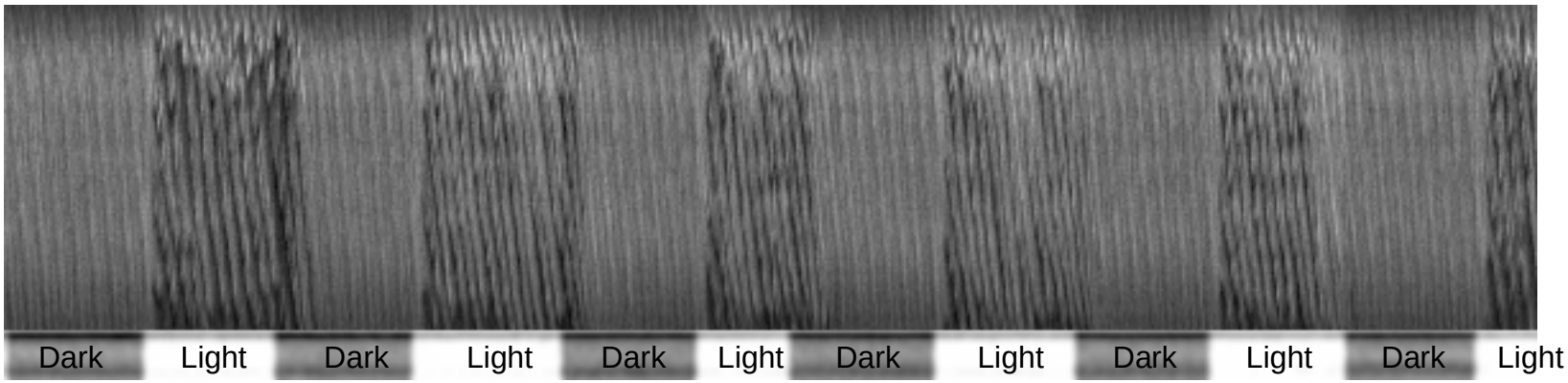
- 500nm monochromatic light beam of 10µm diameter



Selective eye illumination

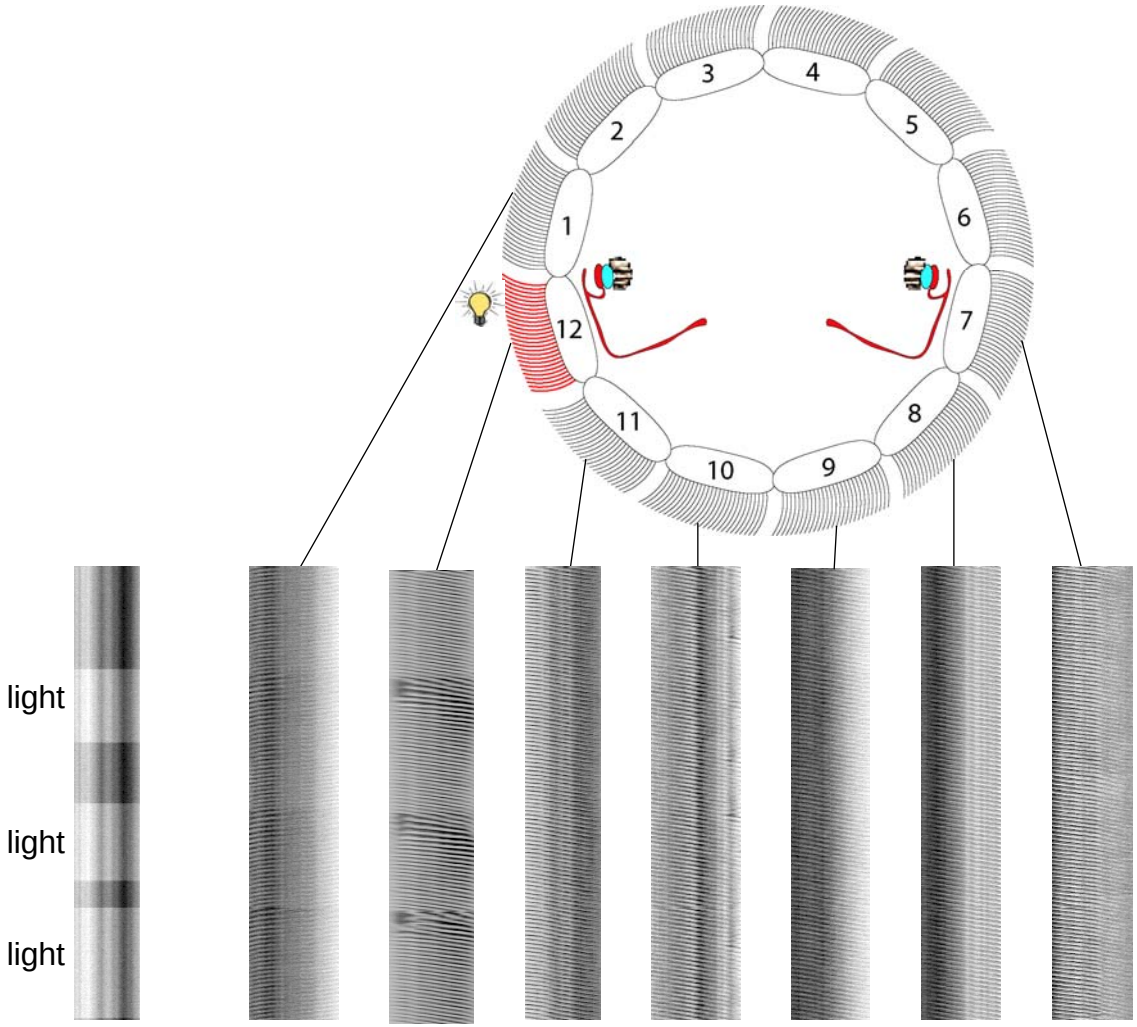
QuickTime™ and a
Sorenson Video decompressor
are needed to see this picture.



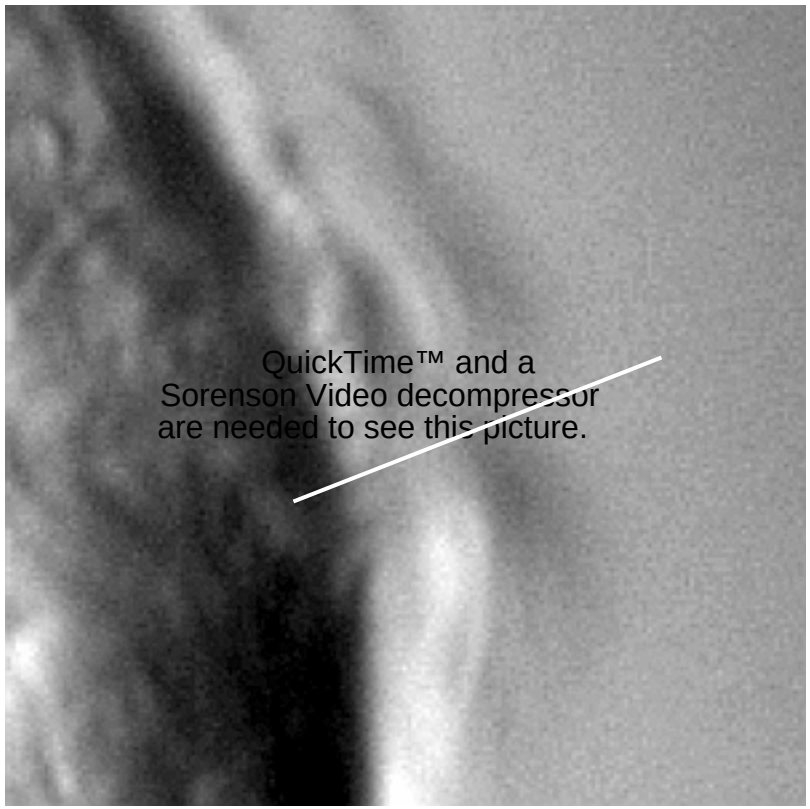


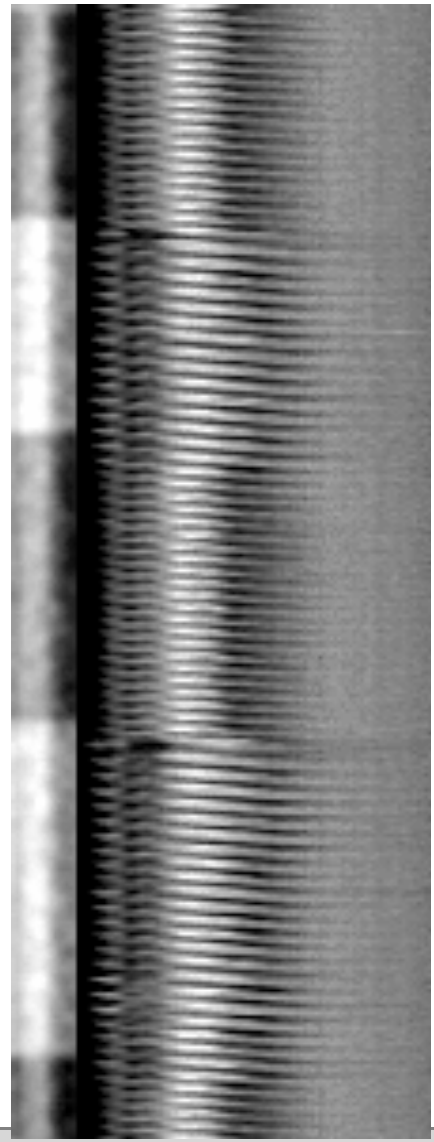
minimum of 215 ms intervals -> fast dark adaptation to detect rapid intensity changes

The ciliated cell closest to the eye changes beating

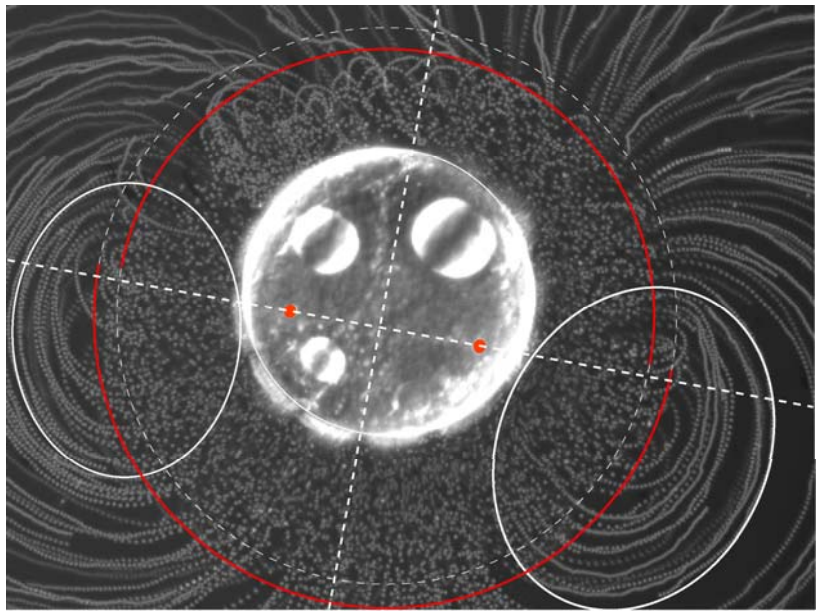


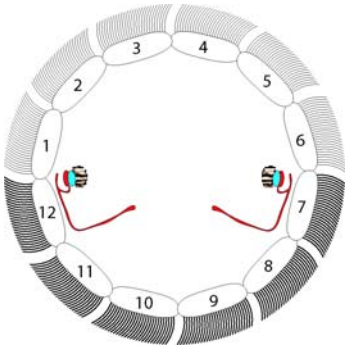
Change in the ciliary stroke pattern

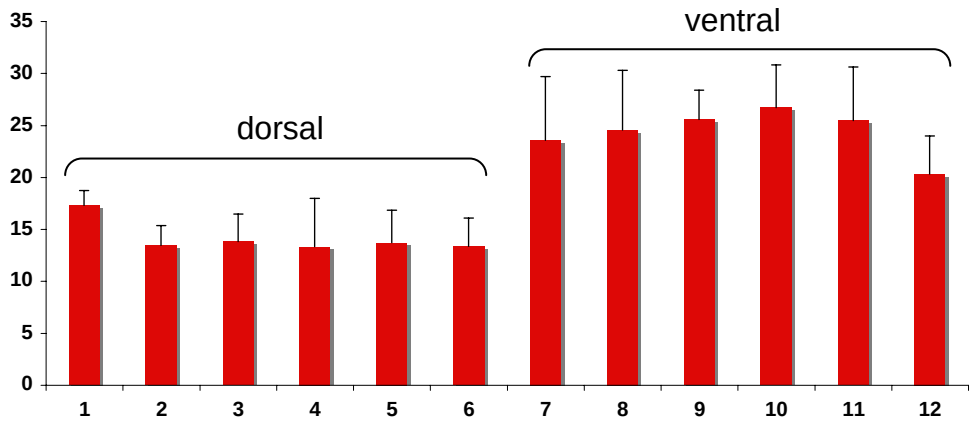




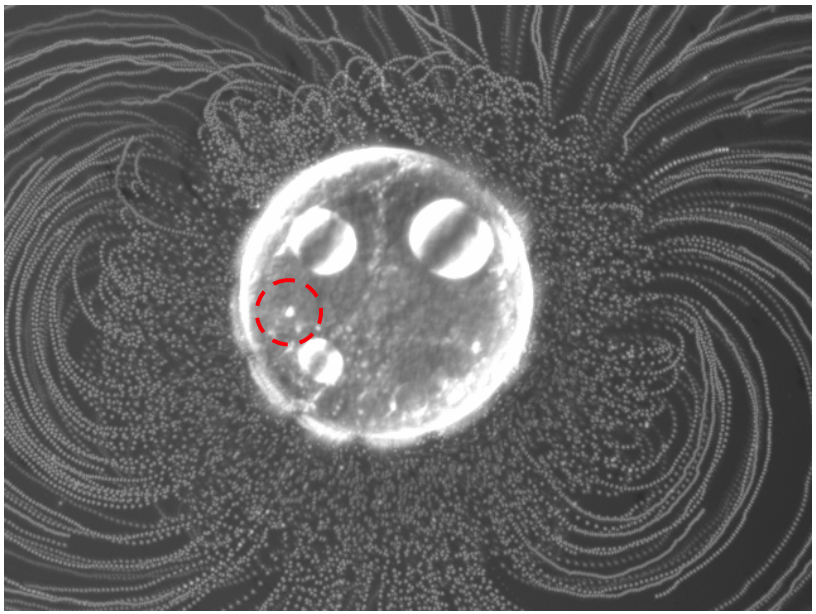
Visualising cilia-generated currents

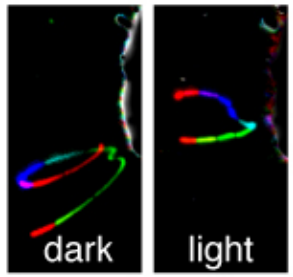


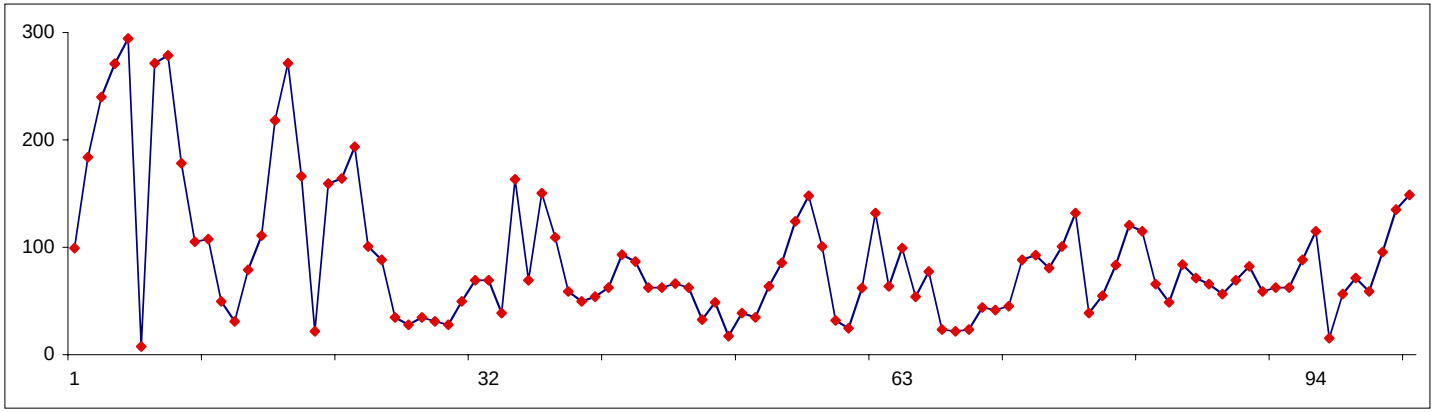




Particles slow down next to the eye upon illumination



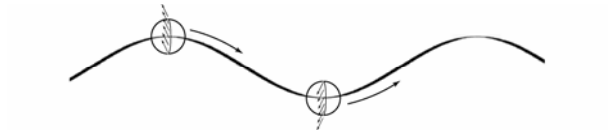


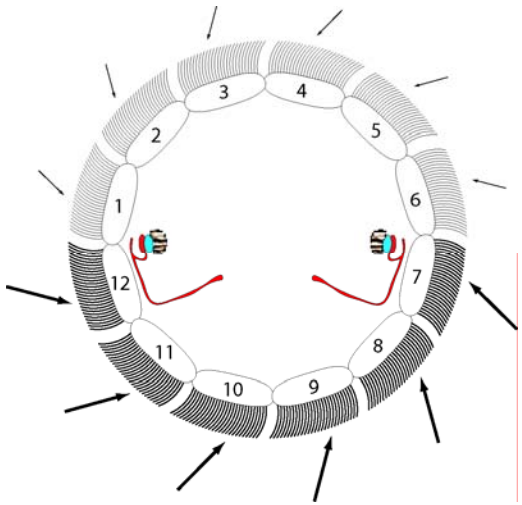


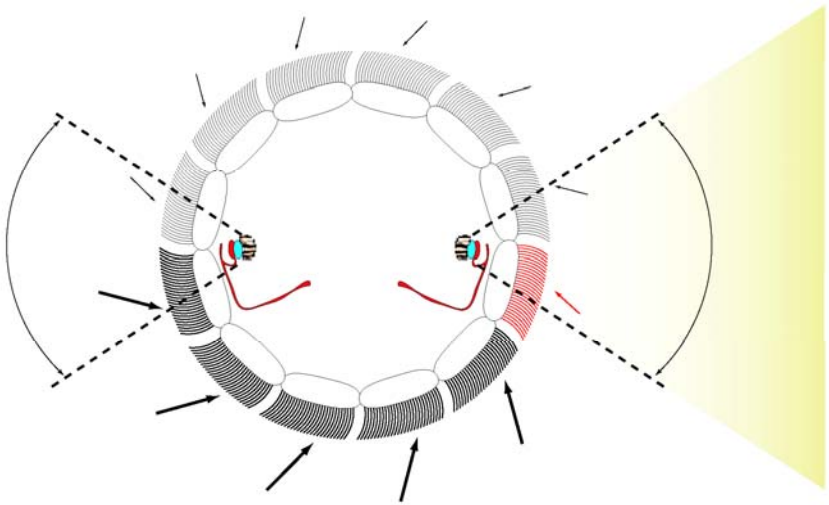
How can this minute change steer helical swimming?

A mathematical model of *Platynereis* swimming

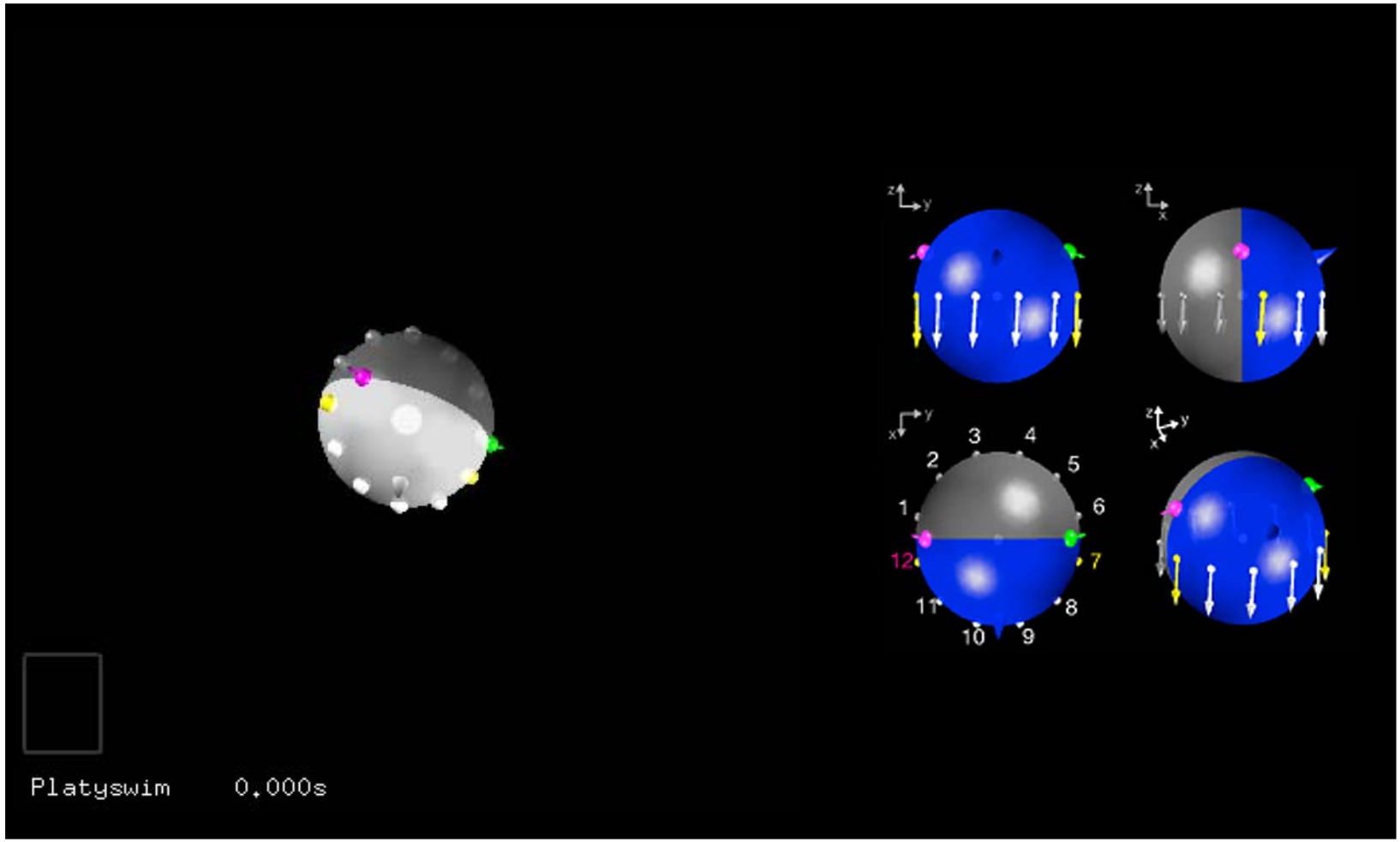
QuickTime™ and a
TIFF (LZW) decompressor
are needed to see this picture.







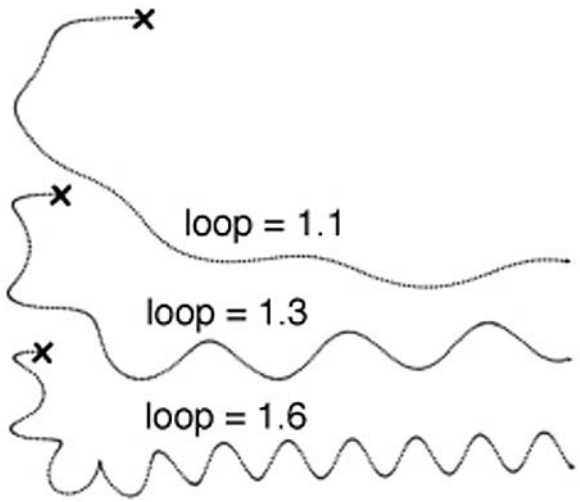
Computer simulation of larval swimming



Computer simulation of phototaxis

QuickTime™ and a
PNG decompressor
are needed to see this picture.

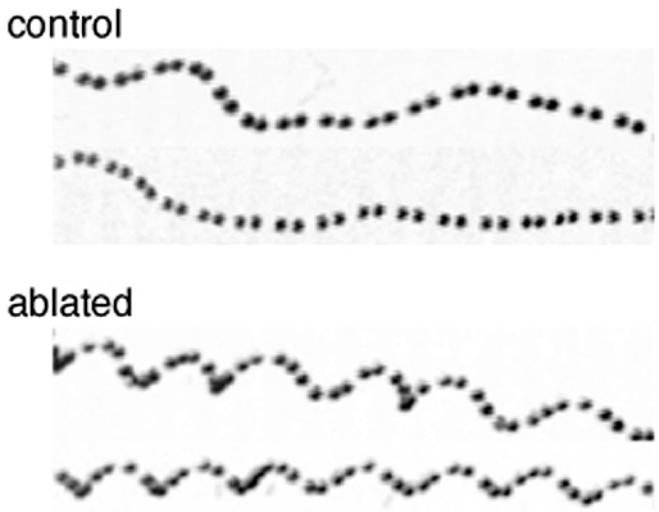
Prediction of the model



Shaving the dorsal cilia

QuickTime™ and a
Sorenson Video decompressor
are needed to see this picture.

Shaving the dorsal cilia



Acknowledgements

EMBL

Benjamin Backfisch
Diana Bryant
Fay Christodoulou
Alexandru Denes
Carmen Doering
Antje Fischer
Daria Gavrouchkina
Keren Guy
Peter Hantz
Gaspar Jekely
Nicola Kegel
Florian Raible
Heidi Snyman
Kristin Tessmar-Raible
Raju Tomer
Maria Antonietta Tosches

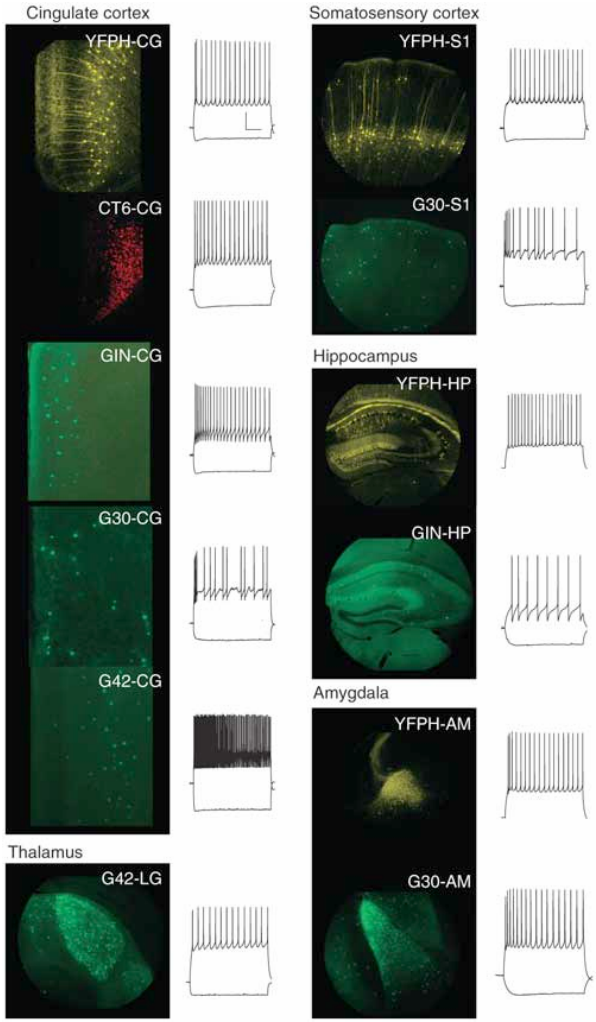
<u>just left</u>
Katharina Willmann
Gaspar Jekely
Carmen Doering

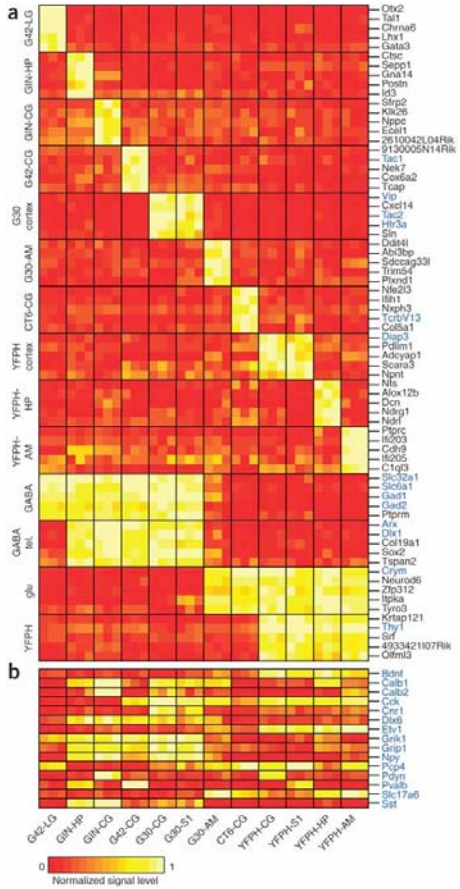
P. Bork , E. Furlong
E. Stelzer, F. Nedelec

M. I. Arnone, Naples
G. Bucher, Göttingen
H. Hausen, Berlin
G. Purschke, Osnabrück
R. Reinhardt, Berlin

G. Balavoine, Gif-sur-Yvette
D. Ferrier, Oxford
Th. Holstein, Heidelberg
M. Akam, Cambridge

From ‘expression profile’ to ‘molecular fingerprint’





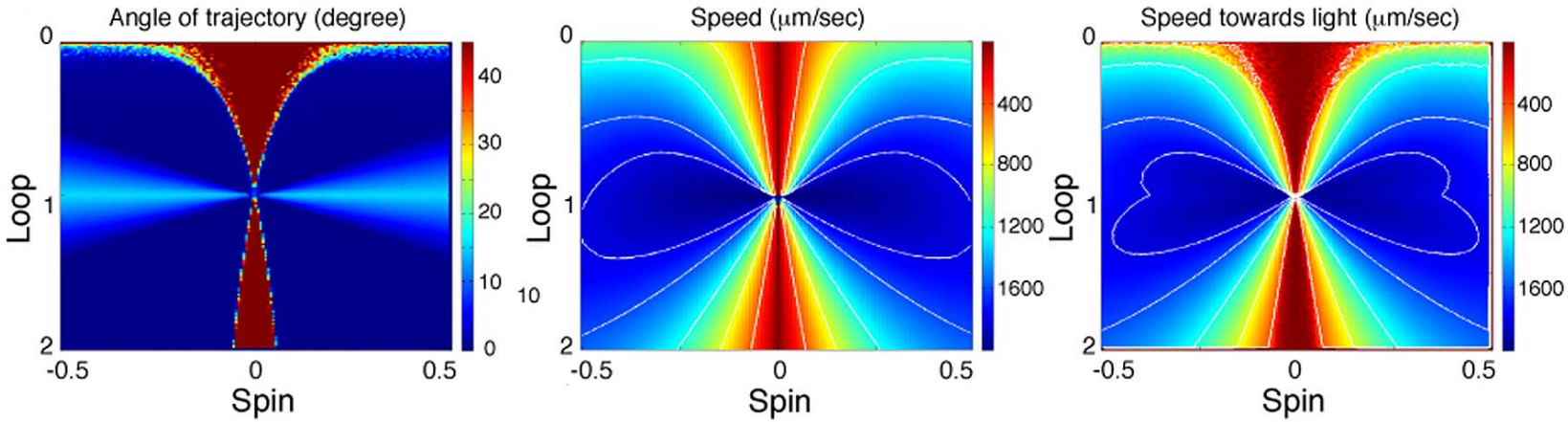
(Sugino et al., 2005
Nelson lab)

Thank you

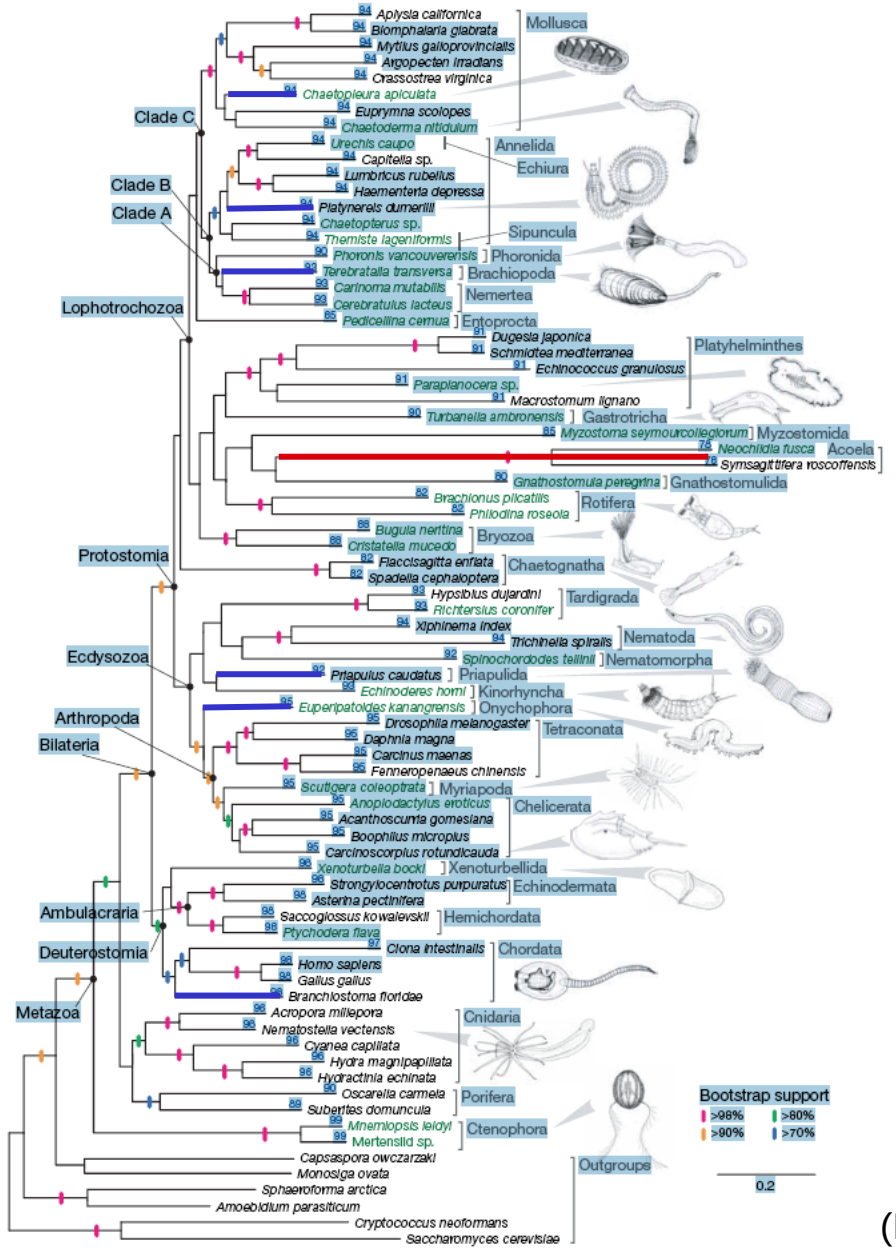
Conservation of cell types: branchiomotor neurons in chordates

Phox2, Tbx20

Dufour et al. 2006

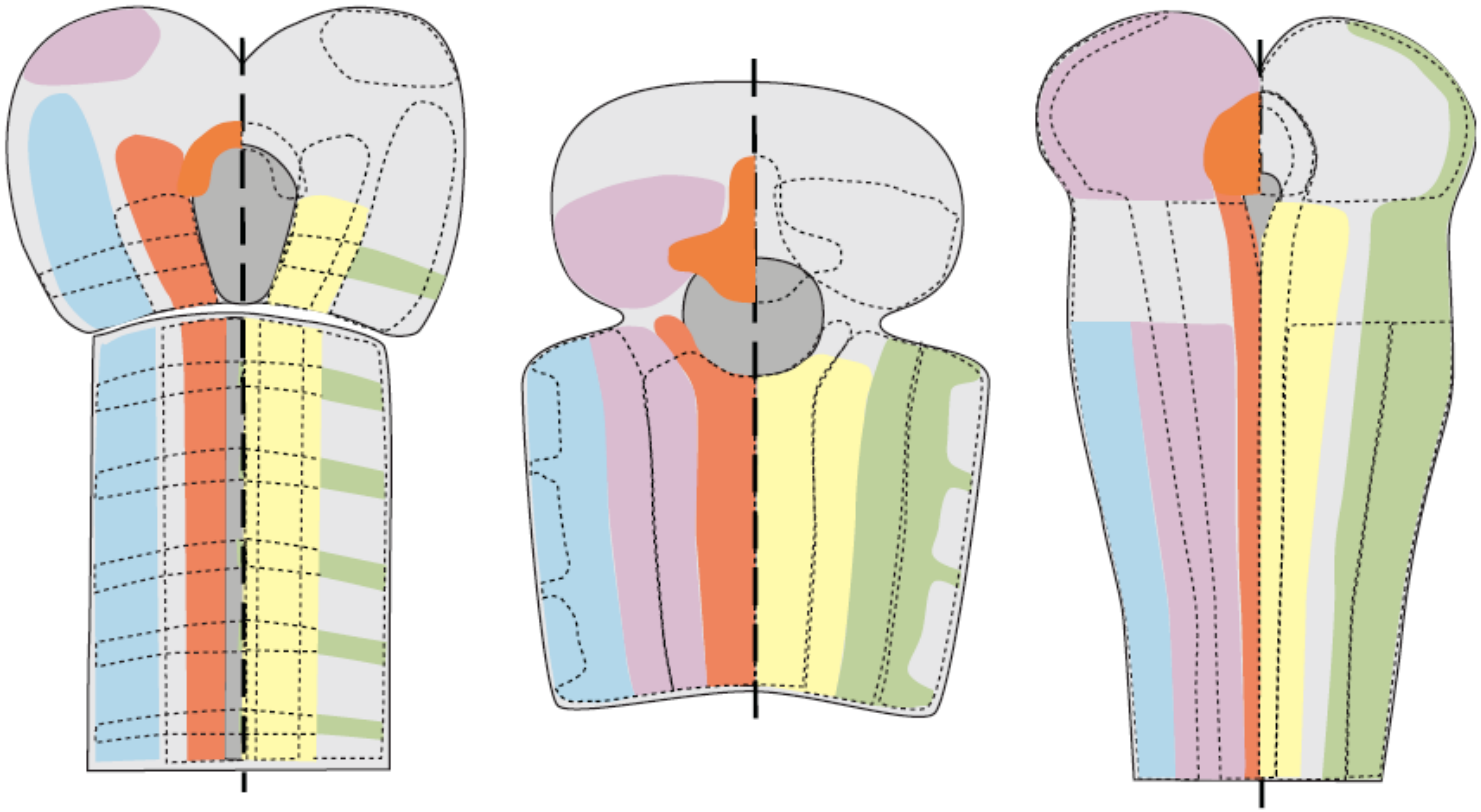

Angle of trajectory (degree)
Loop
Spin
0
1
2
-0.5
0
0.5
40
30
20
10
0
Speed (μm/sec)
Loop
Spin
10
400
800
1200
1600
Speed towards light (μm/sec)
Loop
Spin
400
800
1200
1600

The new bilaterian phylogeny



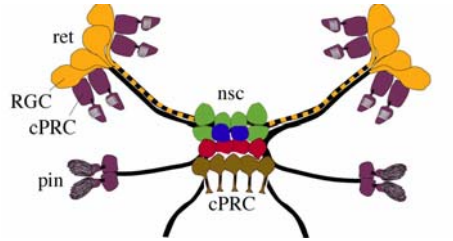
(Dunn et al., 2008)

Conserved mediolateral patterning in Bilateria



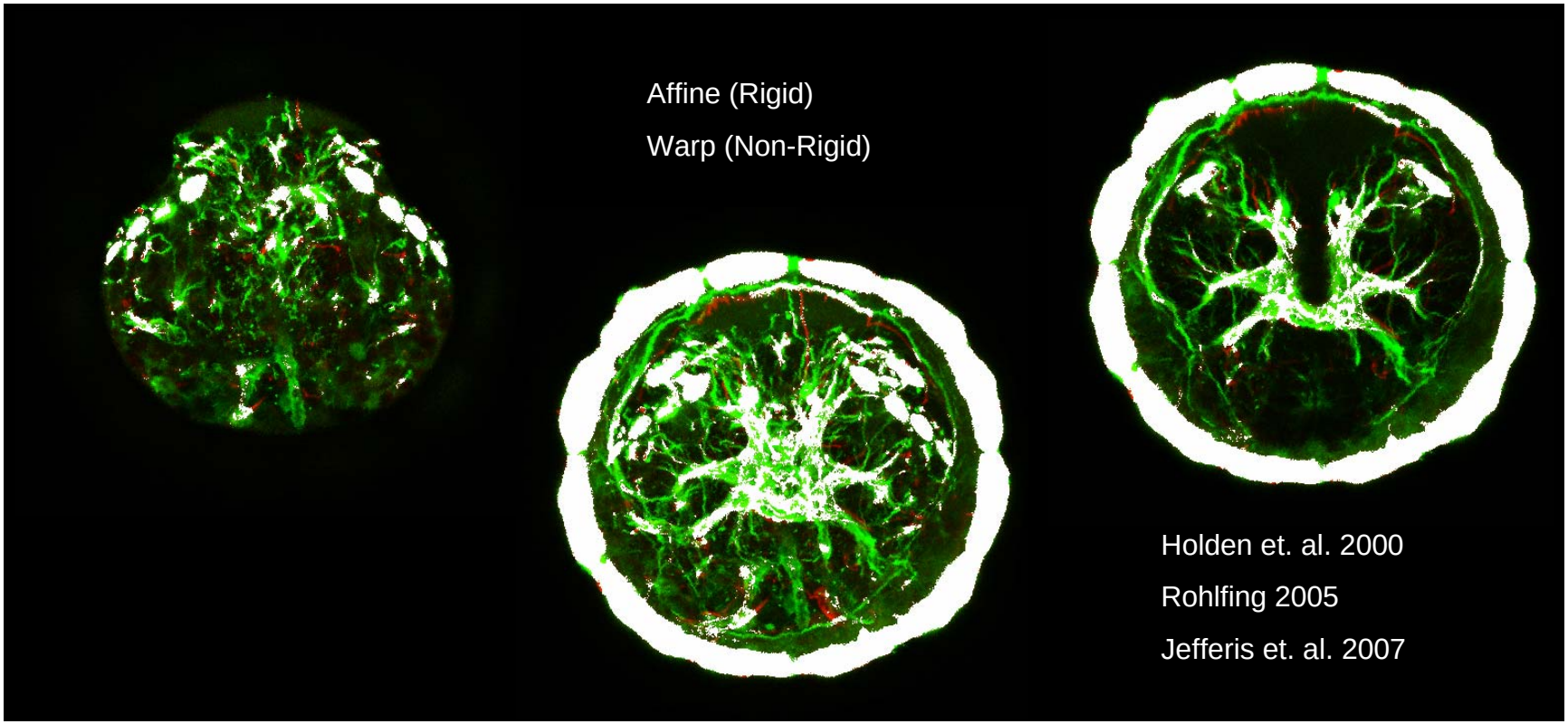
insect polychaete vertebrate

Evolution of photoreceptor cells

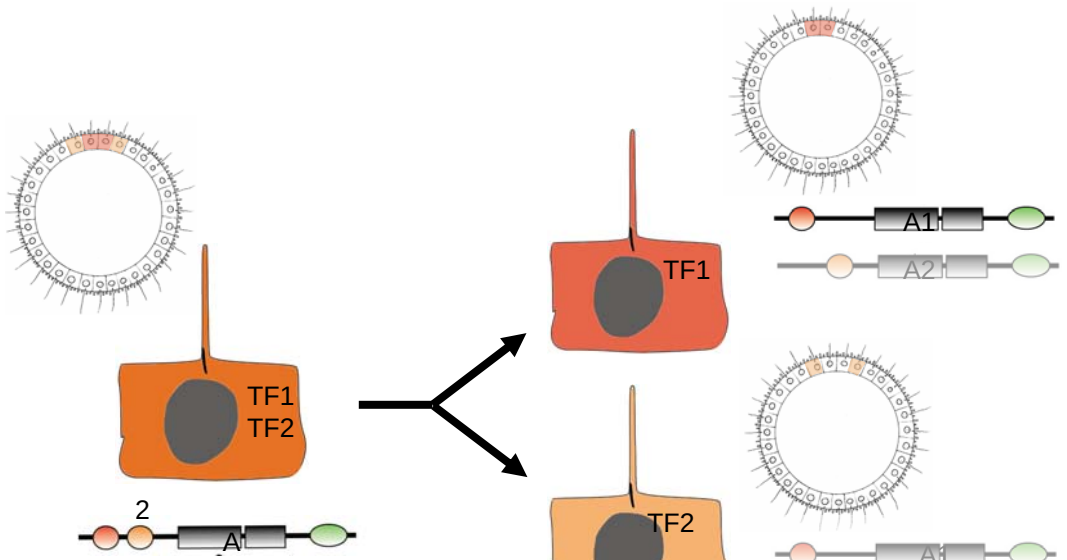


QuickTime™ and a
decompressor
are needed to see this picture.

Image registration by maximizing mutual information



Gene duplication and cell type divergence



(duplication-degeneration-complementation)