

Possible Talk Outline

- Overarching Scientific/Medical/Engineering Context (1-2 slides).
- System Under Study (1-3 slides).
- Key Scientific/Medical/Engineering Questions Addressed (1-2 slides).
- Current Methodologies Employed (1-2 slides).
- System Components (Cell Types, ECM, ...) (2-4 slides)
- Key Component Behaviors (2-4 slides).
- Deficiencies of Current Methods/Approaches, Needs (1-2 slides).
- Results, Focusing on HOW they are Expressed (1 slide).
- Possible Applications of CBO/CBMSL/Repository (1-2 slides).



Guidance

- Think top down.
- Think about how you Describe your Experimental Results/ Simulations to Others.
 - What Components (Cell, ECM...) are Involved?
 - What Behaviors, Morphologies,...are **Crucial** to these Components in your Particular Problem?
 - Start as Generically as Possible.
 - Treat Cells Initially as Black Boxes.
 - Add Detail Hierarchically.
 - **Separate Control (differentiation) from Behaviors?**
 - Stop when you become Quantitative.
 - What Language do you Use to Describe Locations?
 - Do all Components (Cells) of a Given Type Share the Same Behaviors?



Reconstructing multiscale dynamics in animal morphogenesis

Nadine Peyri ras

DEPSN CNRS Gif sur Yvette

Institute for Complex Systems Paris Ile de France ISC PIF

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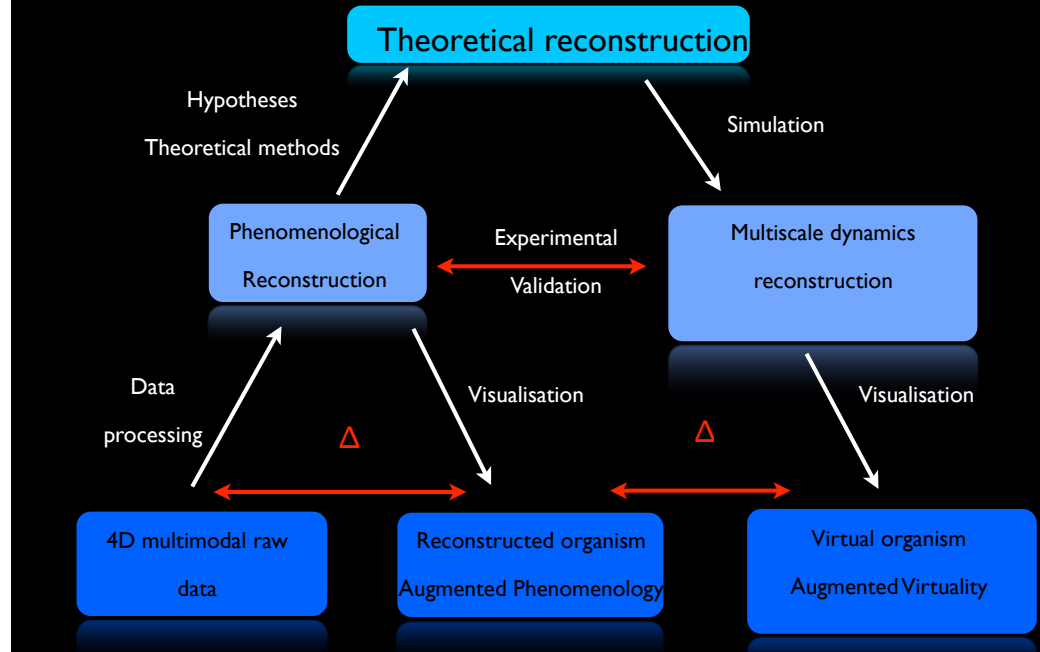


Karlstrom and Kane in zfin.org

automated methods to get the appropriate measurements for reconstructing systems dynamics starting with the cell lineage

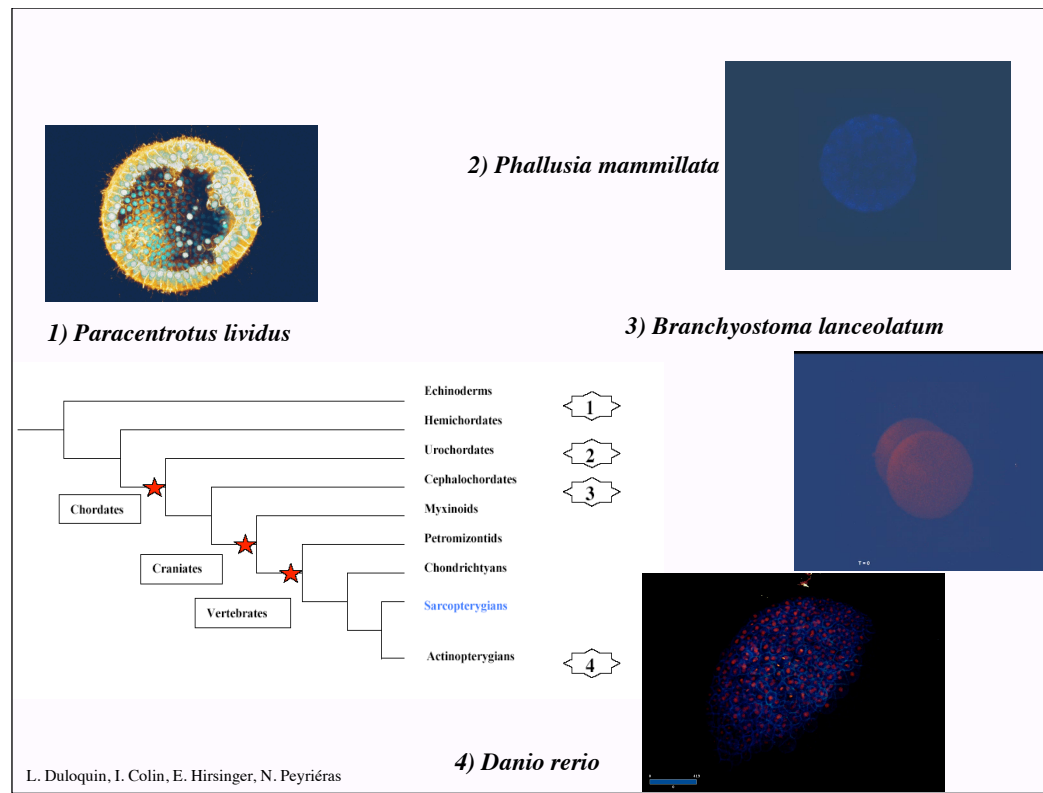
Formal and applied epistemology for the reconstruction of complex systems multiscale dynamics

Paul Bourguine ISC PIF Paris



Reconstructing the Physiome

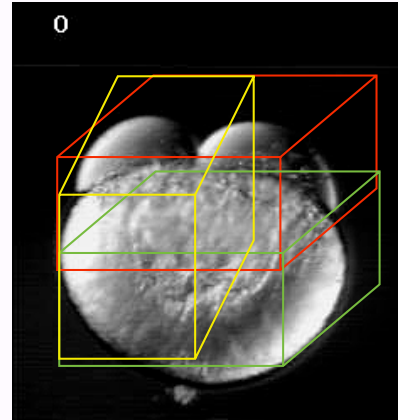
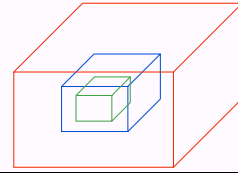
- Reconstruct dynamics observed *in vivo* at the relevant spatial and temporal scales
- Use the cellular level to integrate the different levels of organisation
- Characterize upwards (dynamics of molecular and genetic networks) and downwards causations (biomechanical constraints).
- Identify emerging properties at all scales



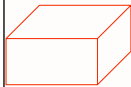
optimizing 4D imaging to get cell position trajectories division shape

Phenomenological and theoretical reconstruction from multimodal imaging

- Molecular and genetic dynamics
- Cell dynamics
- Biomechanical constraints
- Data flow management
- Data processing
- Reconstructions analysis



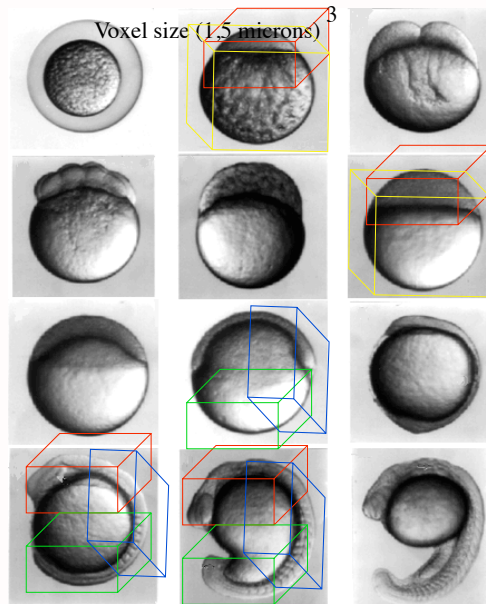
mm size, transparent organisms



775x775x350 microns

(0,4 s/z section < t < 2s/z section)

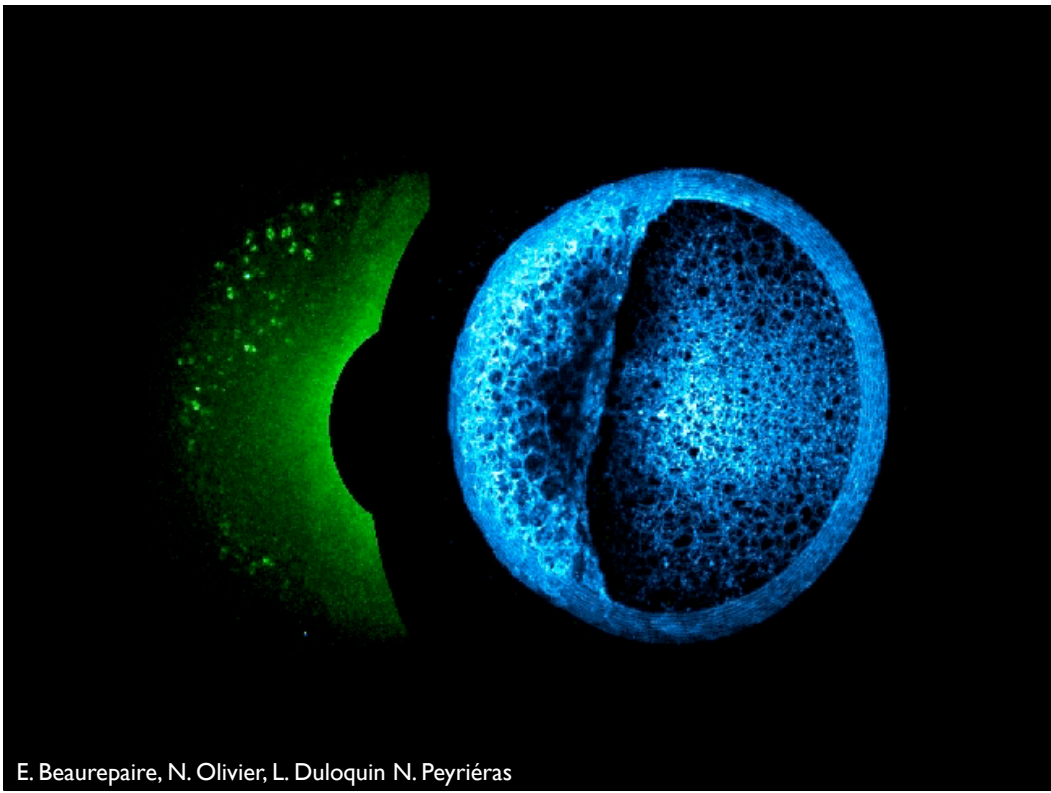
Spatio-temporal tiling MLSM or SPIM/DSL



AP0 or L0-

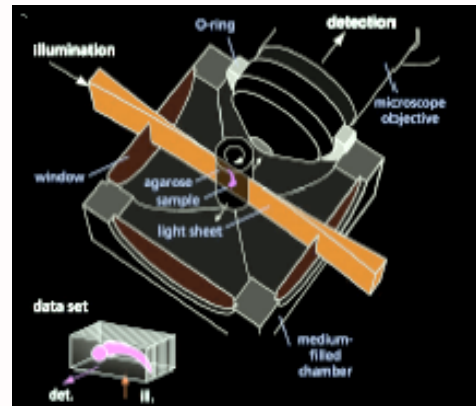
AP4 or L4-

D6 or VP6

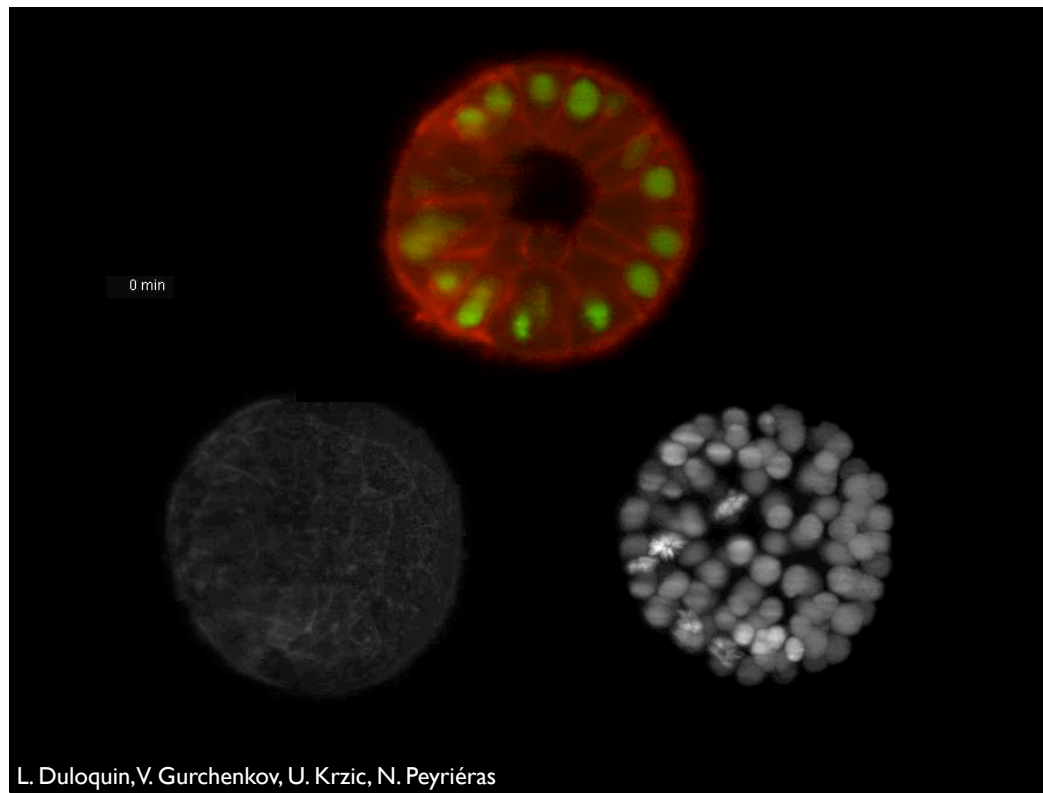


E. Beaurepaire, N. Olivier, L. Duloquin N. Peyri ras

Selective plane illumination microscopy SPIM/DSLIM

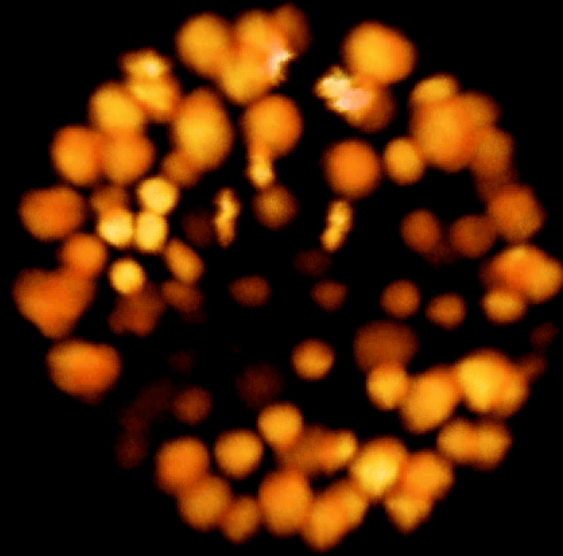


(from E. Stelzer)



60 cellules pendant 20 heures late gastrula

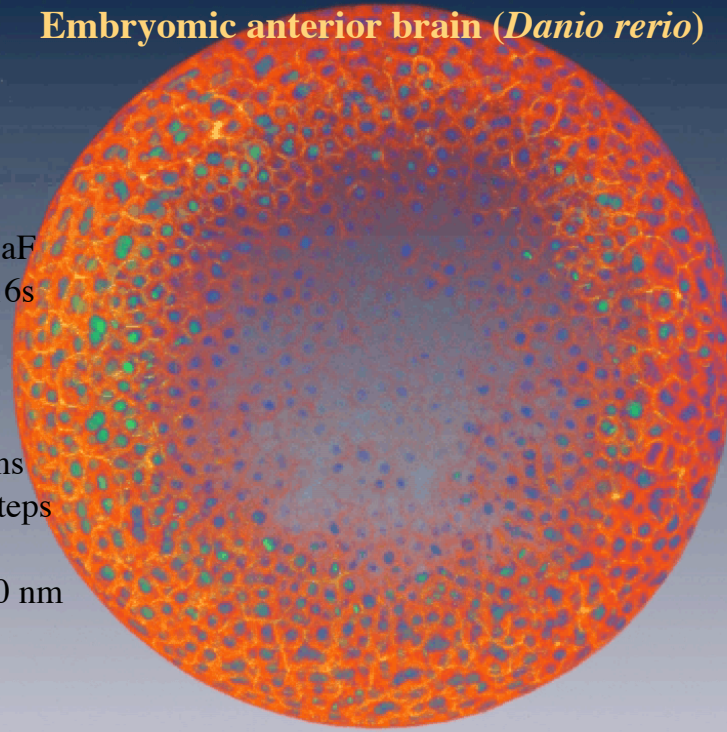
SPIM Multi views fusion



J. Rubio, A. Santos, UPM Madrid

Embryonic anterior brain (*Danio rerio*)

ID 071221aF
sphere to 16s
(1.51)^3
775 in xy
308 in z
205 sections
180 time steps
5'04"
1030 + 980 nm



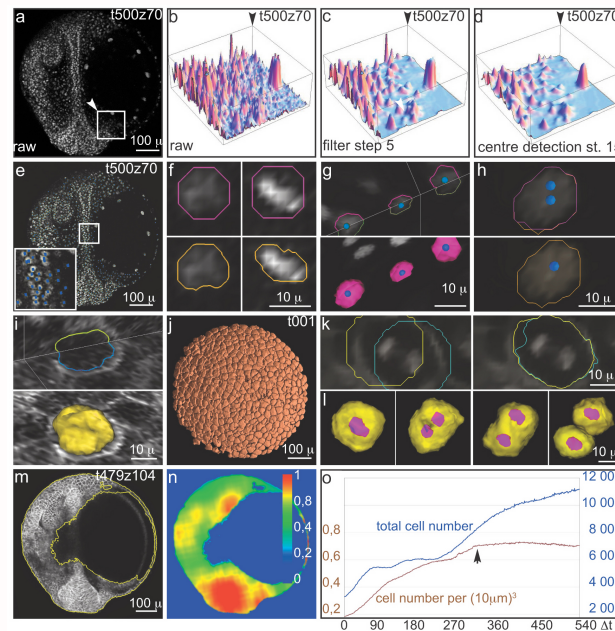
N. Peyri ras

070418a

The phenomenology reconstruction workflow

- **Imaging** at a high resolution (MLSM or SPIM), finding the best compromise (depth, resolution, field size, noise and various arte facts)
- **Processing:** filtering or deconvolution, deblurring, segmentation, tracking = *phenomenological reconstruction* (PDEs, mathematical morphology, estimation maximisation)
- **Measurements and patterns tracking:**
 - Cell shape and behaviours at the micro level
 - Cell motifs at the meso level
 - Clonal origin of organs, compartments and lineage sub-trees
 - Morphogenetic fields as discontinuities in a vector field
- **Data analysis:** finding spatio-temporal correlations, patterns recognition and categorisation,
- **Theoretical reconstruction:** dynamics modelling

Reconstruction chain of algorithms



Geometric
Mean
Curvature
Flow
Flux-Based
Level Set
Center
Detection

Subjective
Surface
Technique

K. Mikula et al., Sarti et al.

Computational Image Processing Strategies, Methods and Tools

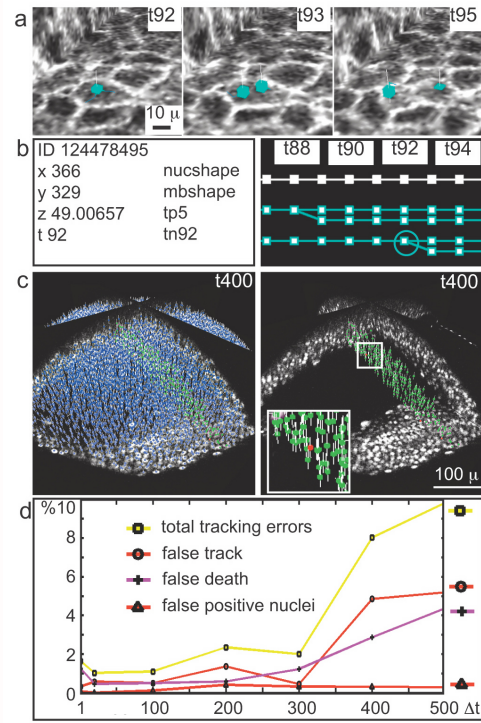
(PDEs) that present the remarkable adaptive properties of the mammalian visual system.

— image filtering— scalar intensity functions in grey level scale. We selected in the geometrical nonlinear partial differential equations family, the so-called geometric mean curvature flow (GMCF) in the level set formulation that appeared to be the most appropriate model [KMPRS]. These flows perform nonlinear multiscale analysis and are contrast invariant.

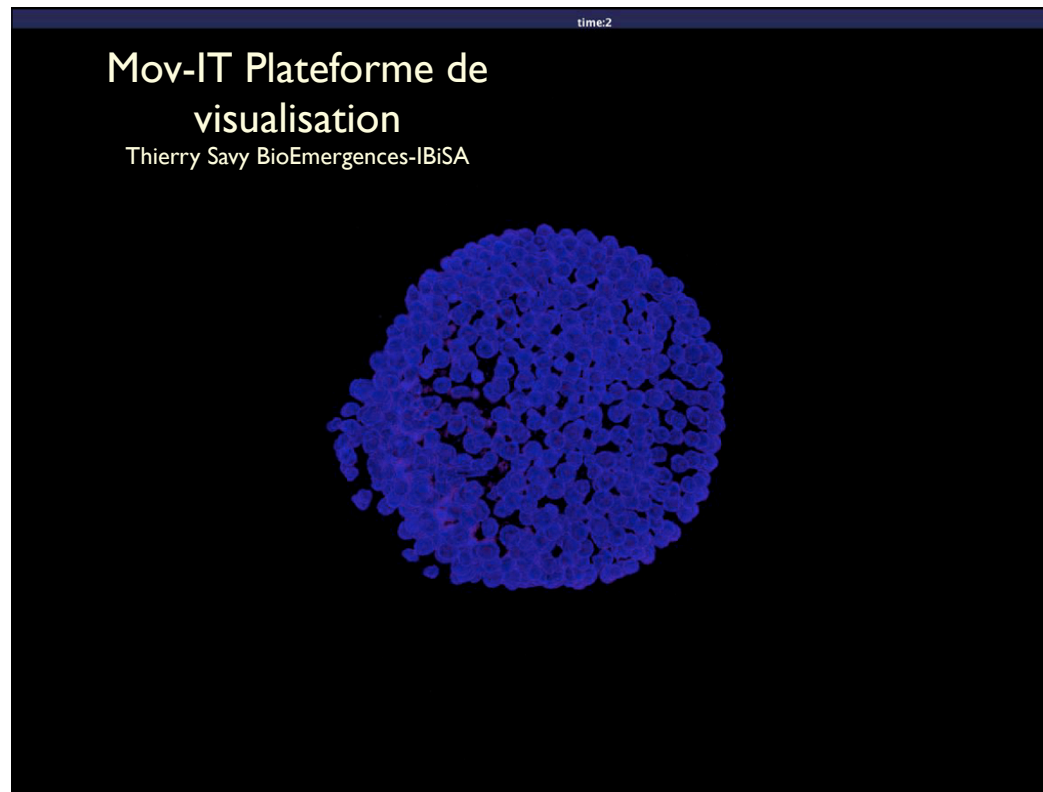
For the second step — nuclei center detection — we use another nonlinear multiscale strategy based on PDEs, called flux-based level set center detection (FBLSCD). The principle of this process relies on the fact that all visible objects in the image can be seen as humps of relatively higher image intensity. This multiscale method makes the hump decreasing until a stopping condition adapted to the rest of the algorithm chain.

For the third step — the cell nuclei segmentation and cell membrane segmentation — the approximate cell centers represent the point of view (the gaze) from which a point of view surface is constructed. The evolution of the point of view surface with respect to a metric induced by the image tends to a minimal surface in a Riemannian manifold representing the segmentation of the cell. This subjective surface technique (SST) has been introduced in [SMS1, SMS2, SC] to perform perceptual completion in mammalian vision and here it is applied to cell nuclei and membrane segmentation for its ability to fill missing information in the image [CMSSg, Z1, Z2, MPRS]. The numerical technique for implementation is based on co-volume methods [MSSg].

Reconstruction validation



B. Lombardot, T. Savy and Embryomics EC project consortium



070426d 15 20 hpf 57 seconds ?? (63 1,2 oil)

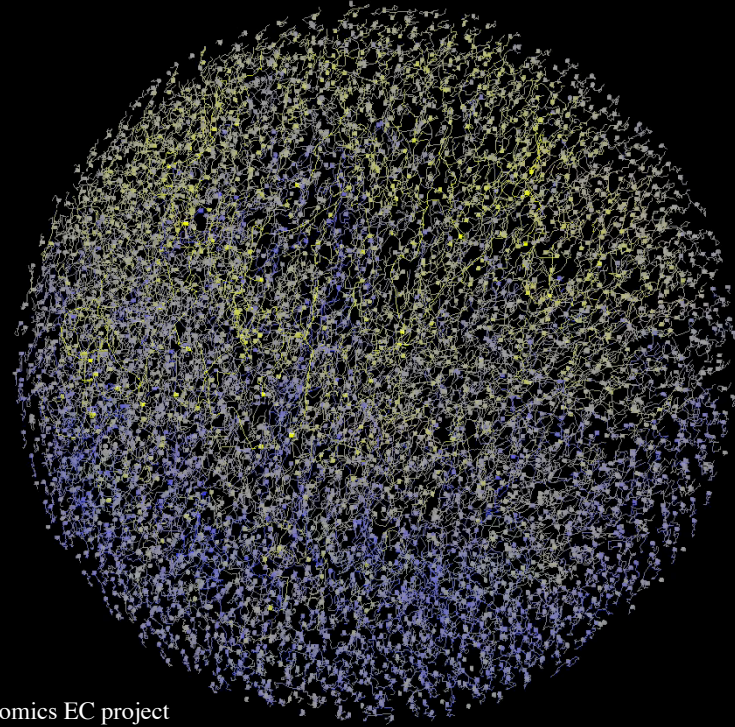
the tools reconstruction visualisation analysis

raw reconstructed

cell position trajectories division

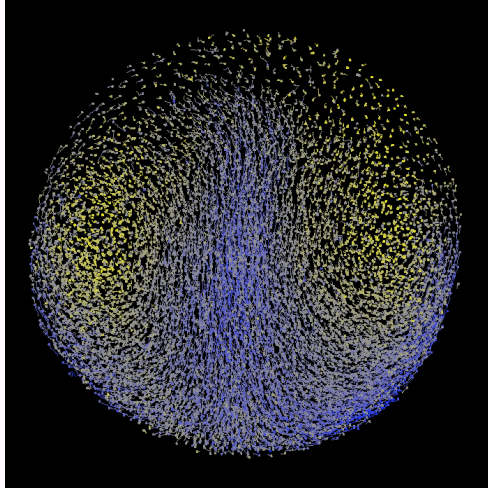
cell selection here endomesodermal part in green gut SMCs red PMCs pink based on the epithelium mesenchymal transition



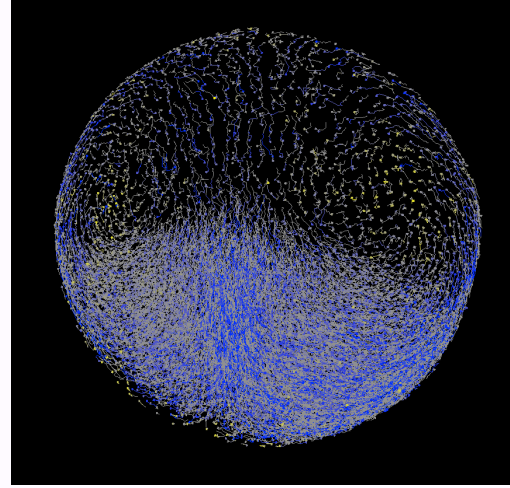


T. Savy Embryomics EC project

Measure, categorize, compare,

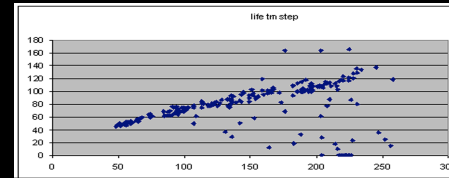
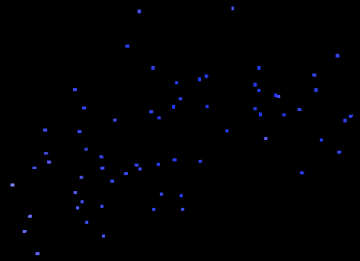


WT

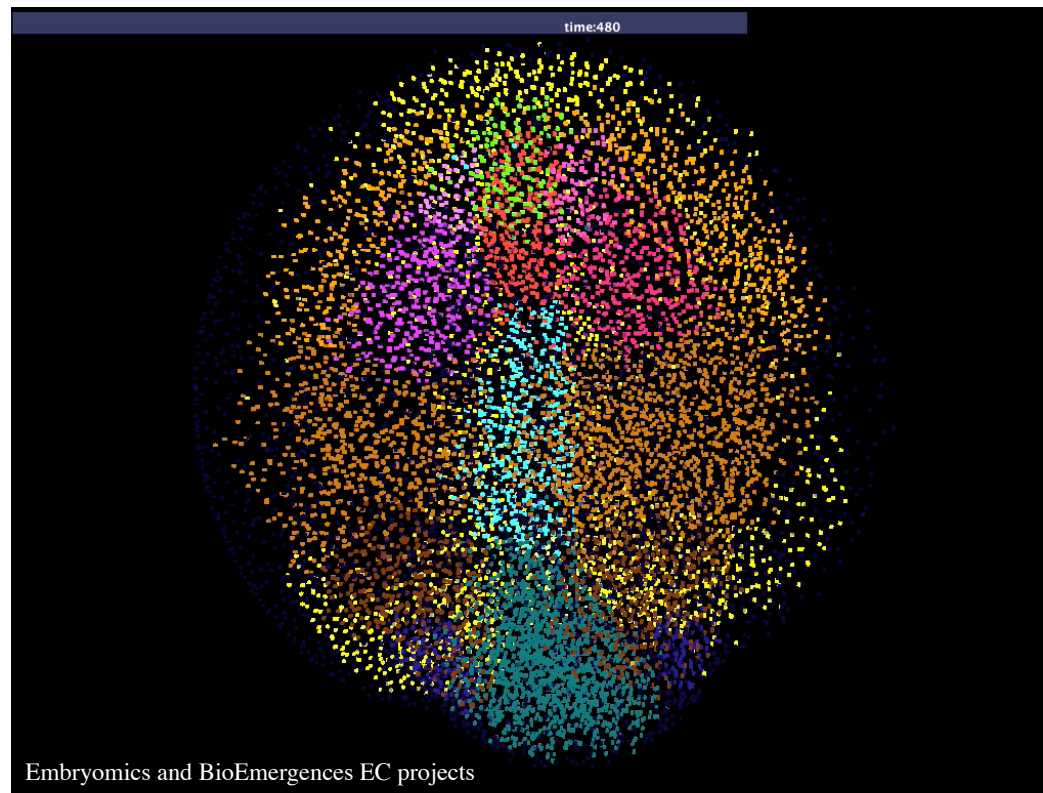


Mutant *zoep*

clonal analysis and spatio temporal correlations

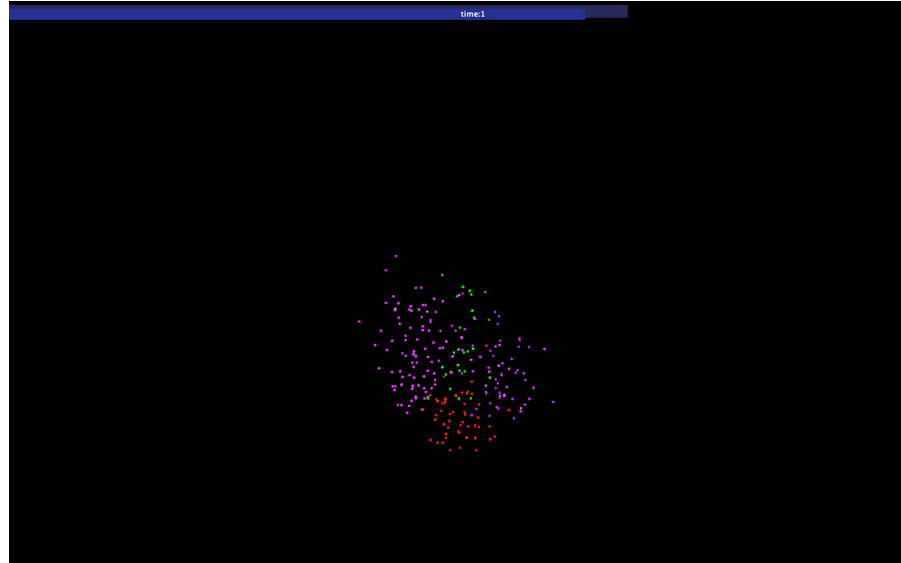


Embryomics and BioEmergences EC projects



- Filtering — geometric mean curvature flow (GMCF)
- Nuclei center detection — flux-based level set center detection (FBLSCD).

**Fate mapping at the midline: eye field
separation is achieved before the onset of
gastrulation**

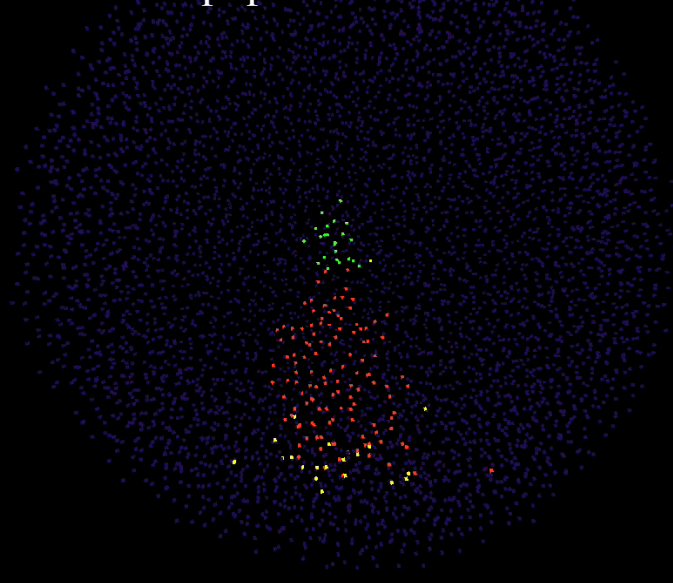


Midline cells do not contribute to eye formation

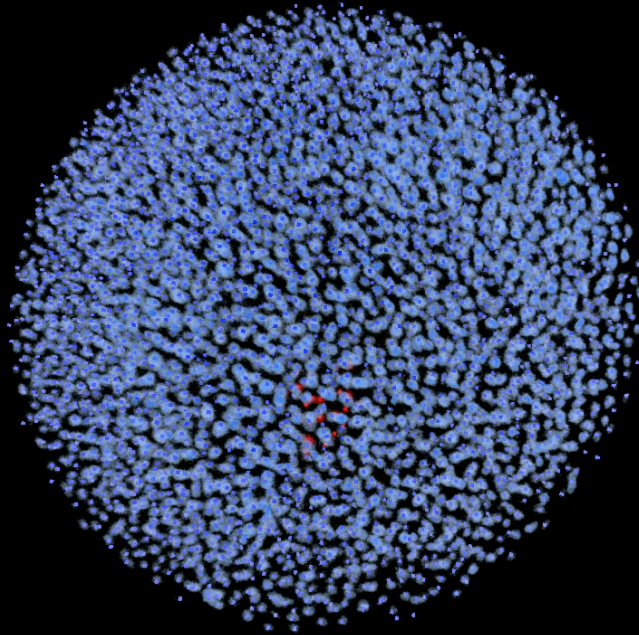
time:1



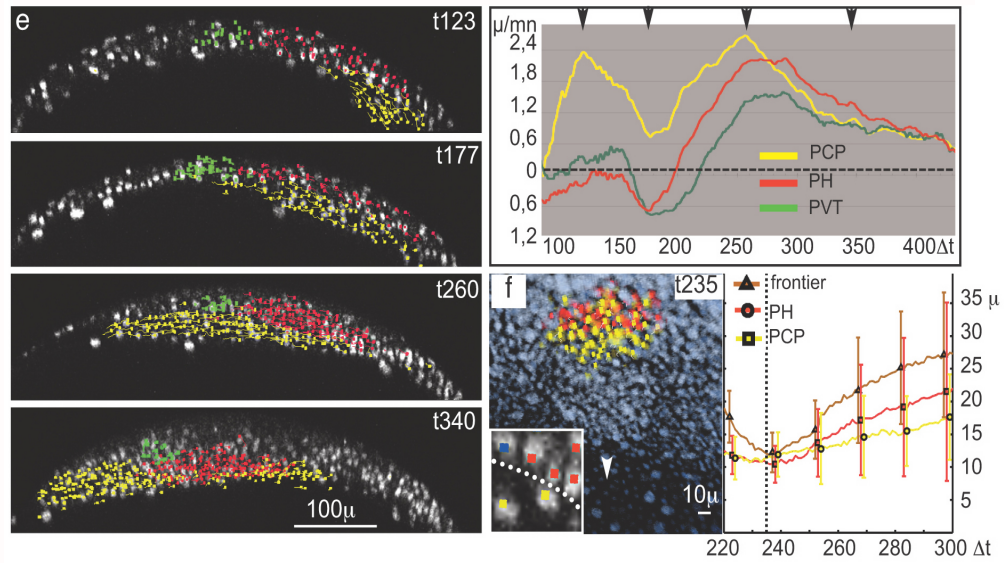
Prechordal plate neuroectoderm interaction: a matter of cell populations relative movements

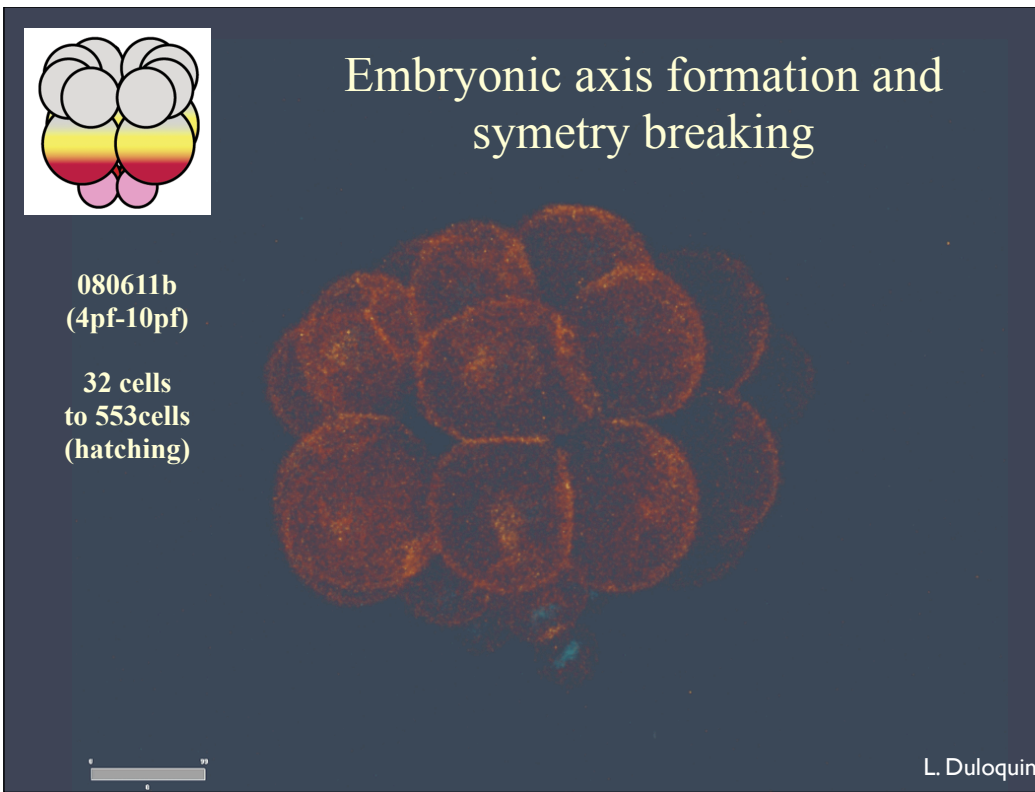


Cell neighbourhood at the midline: Presumptive ventral forebrain and PCP are sliding on each other



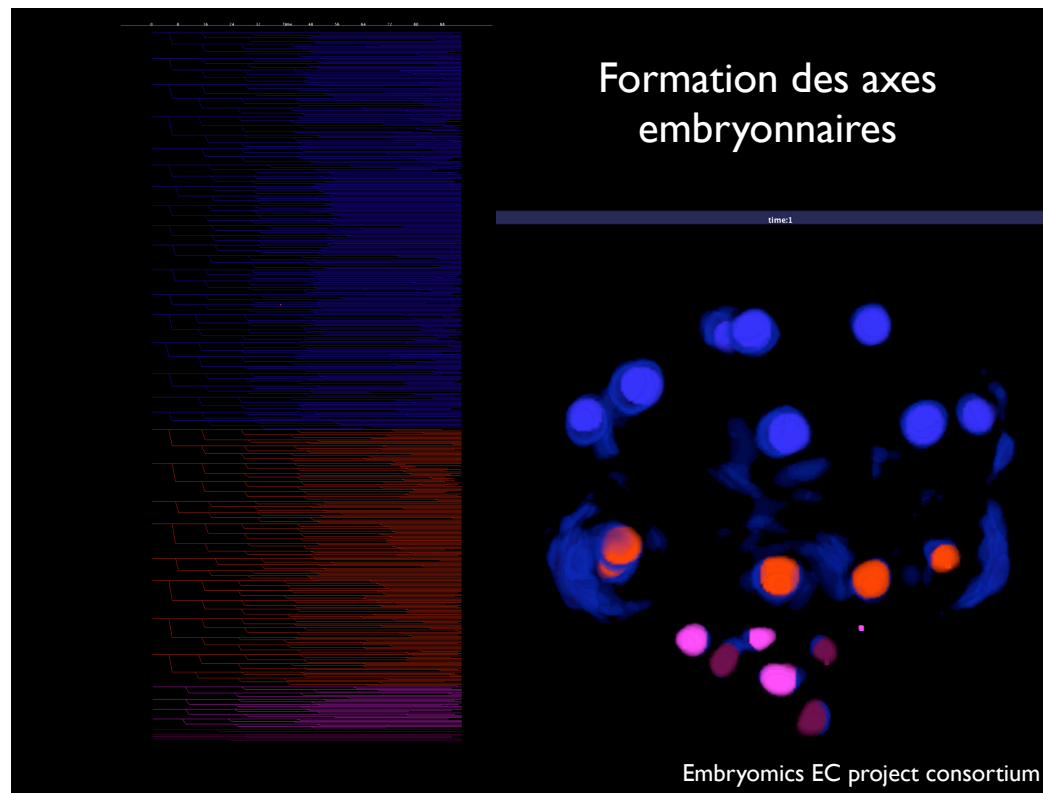
From measurements to biological insights: *in silico* experimental embryology





080611b 3 minutes 27 seconds

Typically, embryos are time lapsed for 5 hours of development. An image data set is made of a volume of 100 sections (slices) encompassing the whole embryo and this volume is recorded every minute.



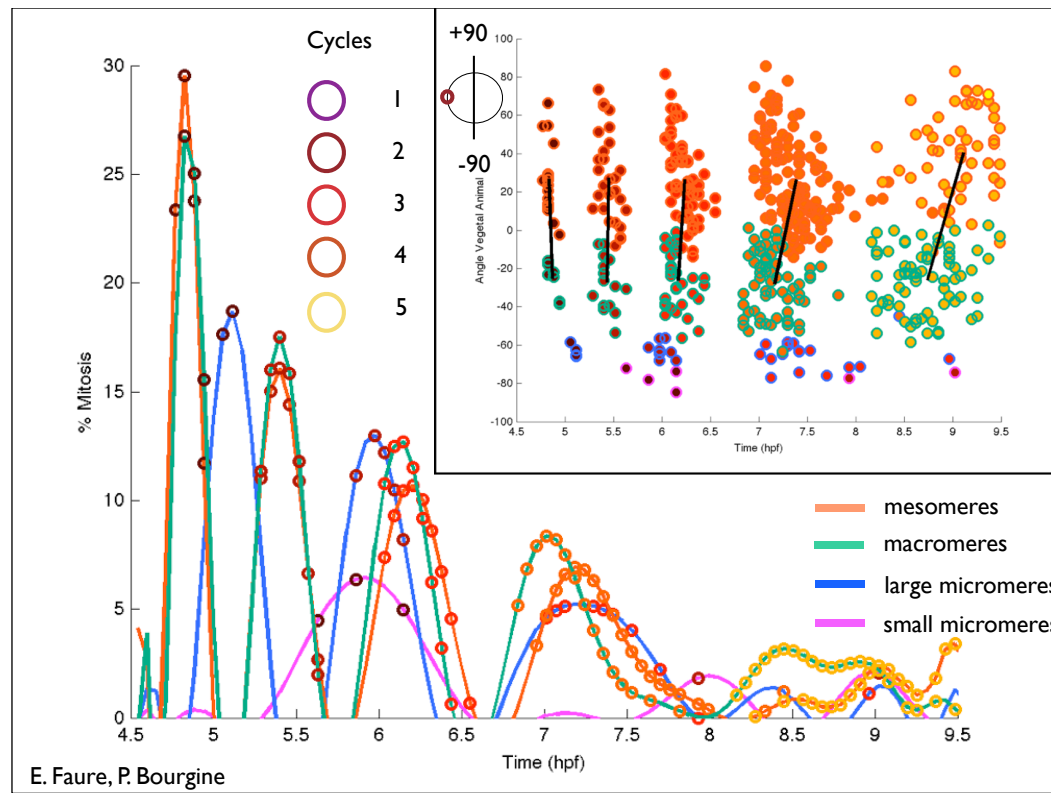
toujours 080611b

small micromeres (in dark) large macromeres in pink both 3D rendering and raw data

macromeres in orange

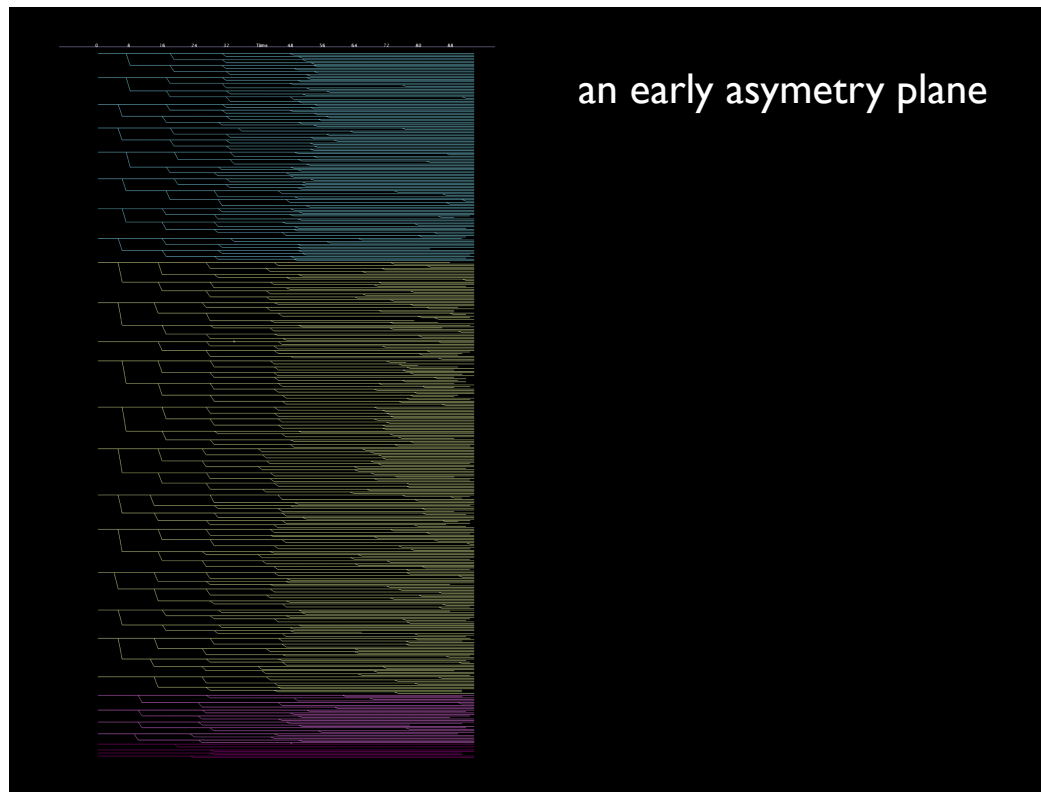
mesomeres in blue

early features such divisions synchrony and its correlation with morphogenetic events



E. Faure, P. Bourguin

080611b (two other data sets) from the 32 cell stage, mesomeres and macromeres continuously desynchronize
 5 waves (colours) small and large micromeres known measurable comparable
 inset: mitosis position expressed as an angle with the animal vegetal axis
 average of macromeres and mesomeres population
 -90 +90 angle Phi

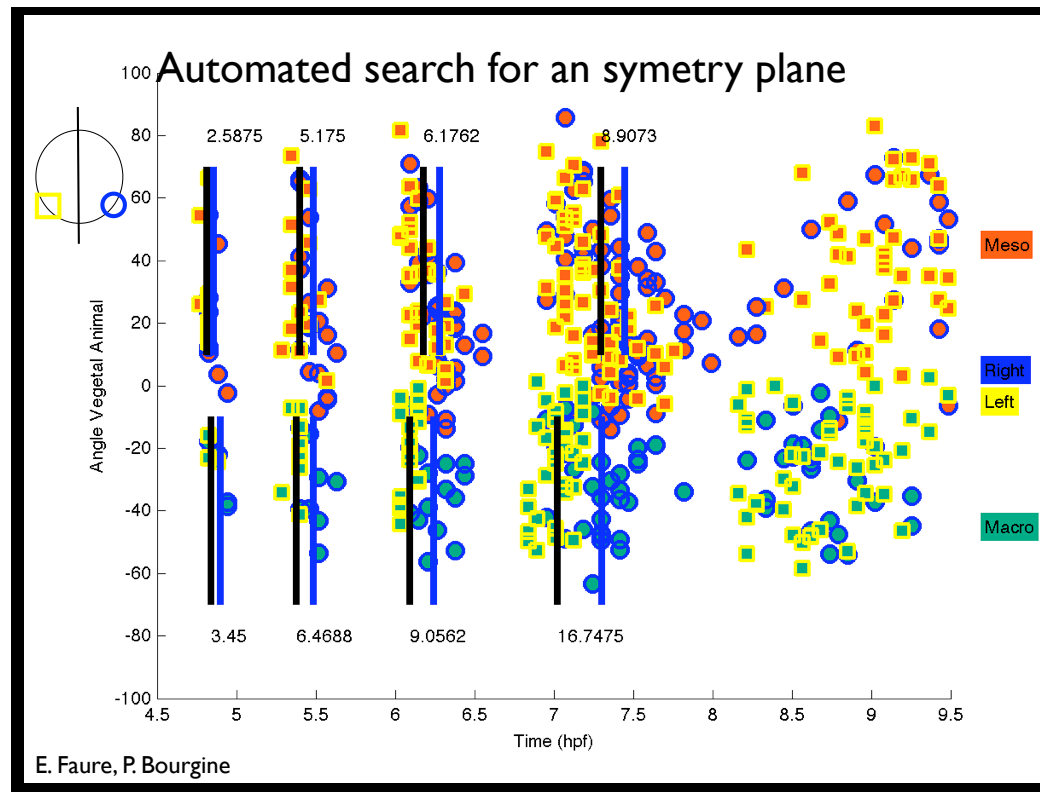


oral en jaune

another way to look at it with one more division on one half

track it back at the 32 cell stage at the time of asymmetric expression of Nodal

Still speculating either late evidence through the lineage or early evidence through transgene expression

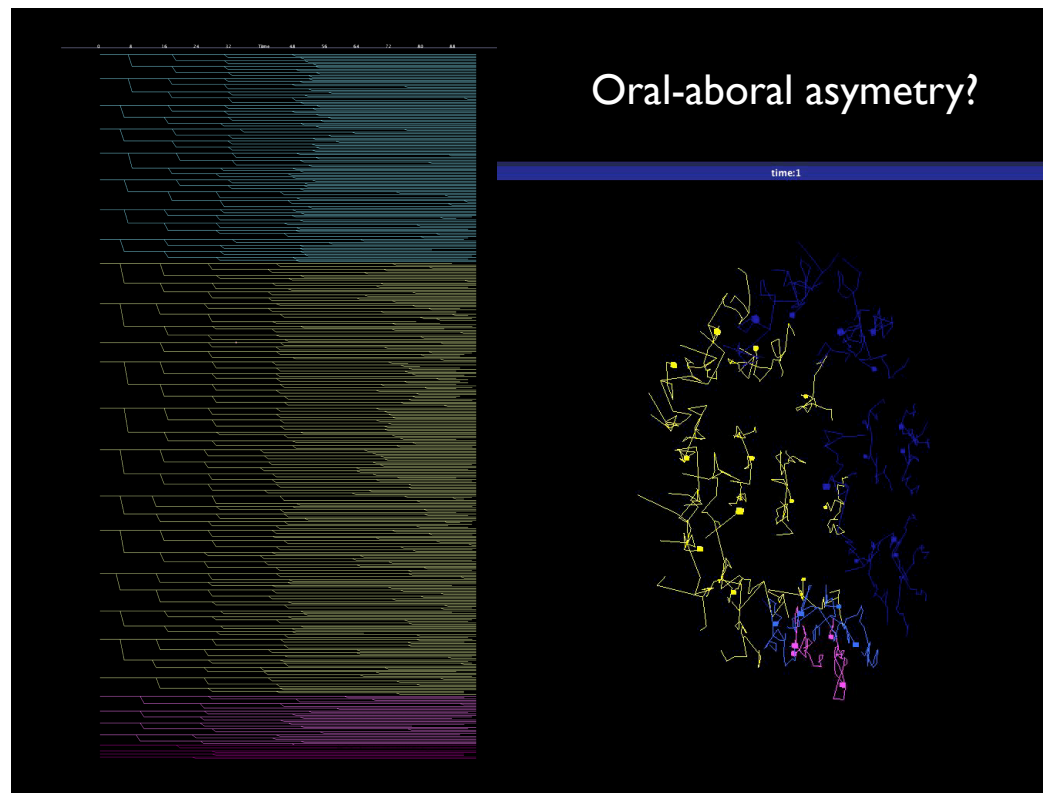


5 vagues

decalage gauche droite qui croit de 3 a 16 minutes

plus prononce dans les macromeres

barre indique le temps moyen de division



oral en jaune

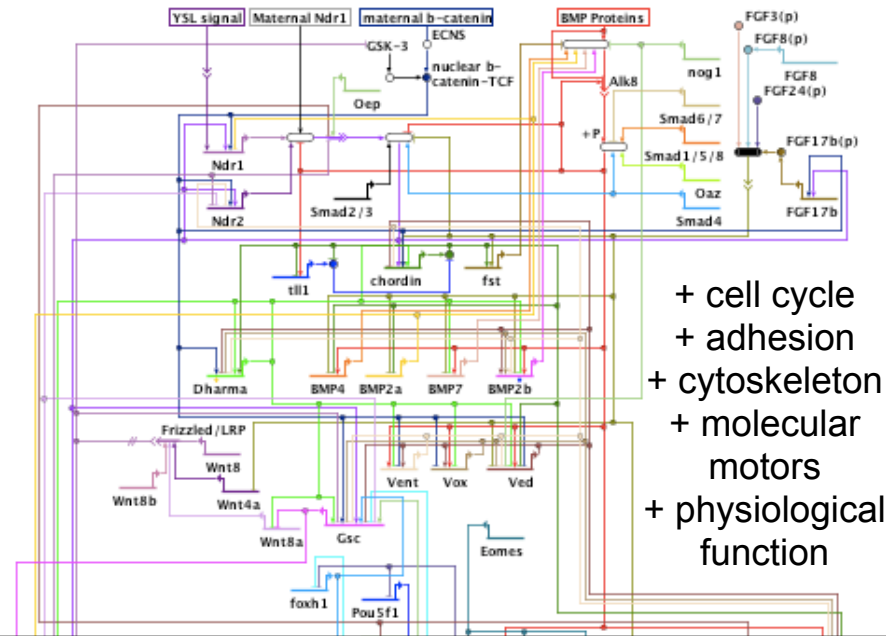
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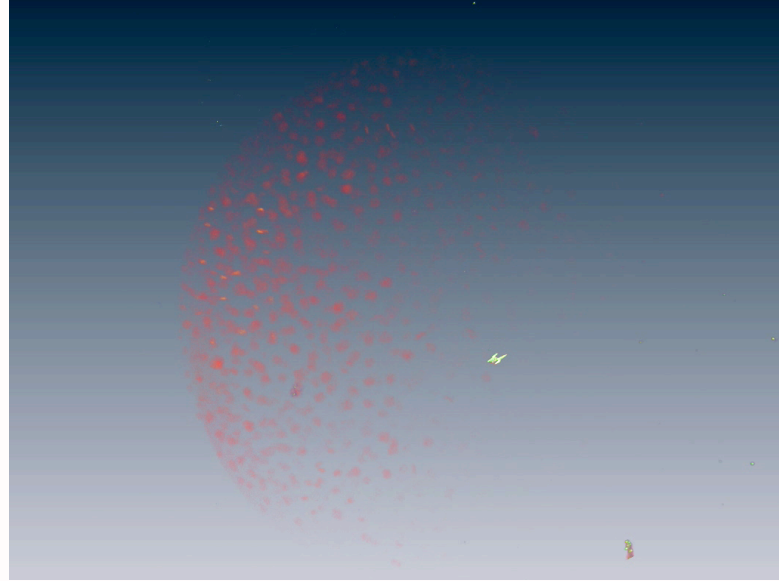
Still speculating either late evidence through the lineage or early evidence through transgene expression

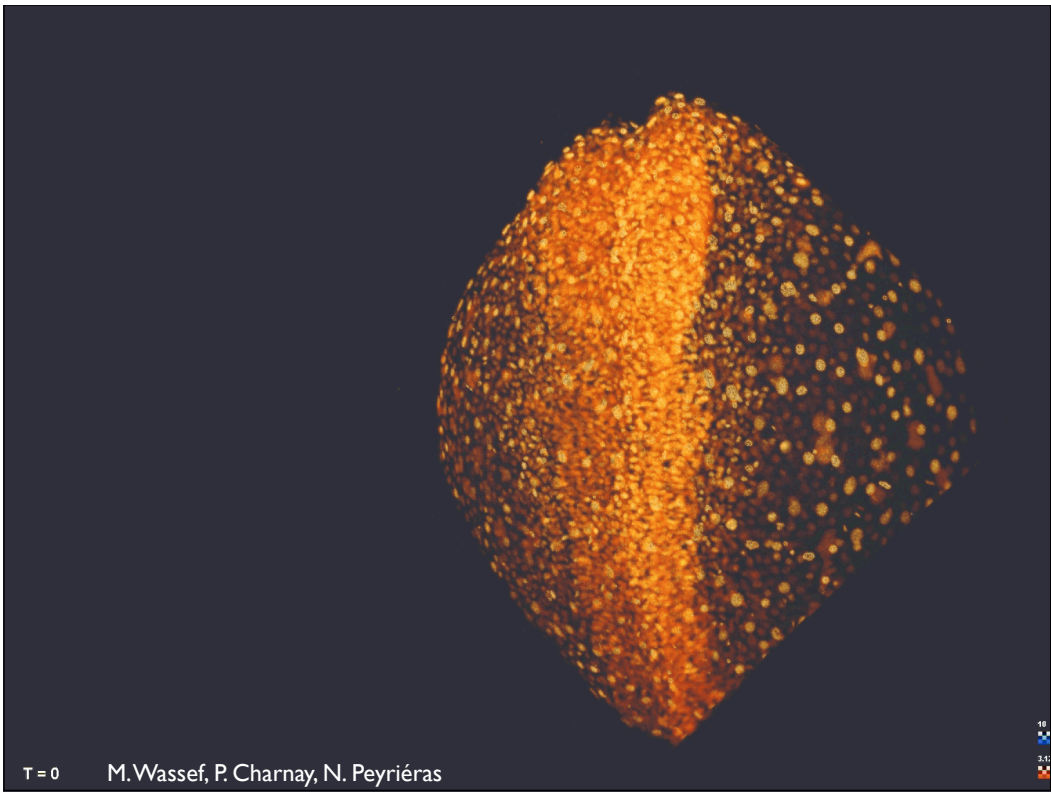
Molecular and genetic networks: from architecture to dynamics

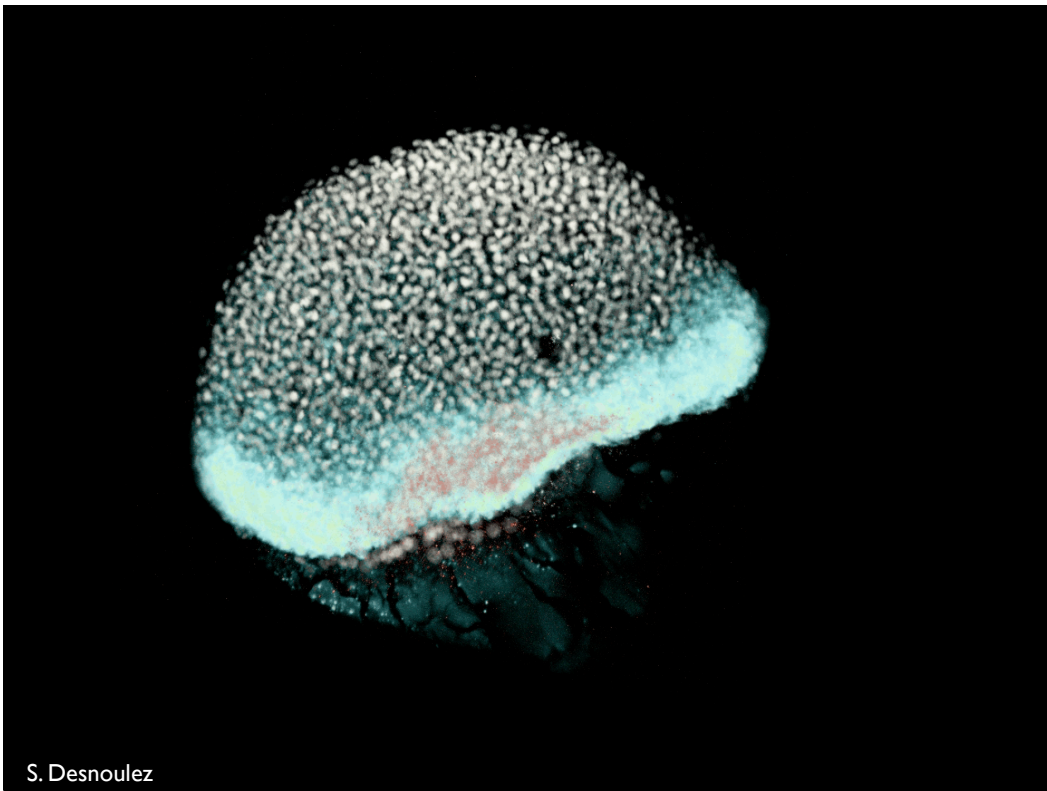
BioTapestry Yuh's lab



Integrating molecular and cellular dynamics: how does gene expression regulation fits with cell lineage and cell fate?

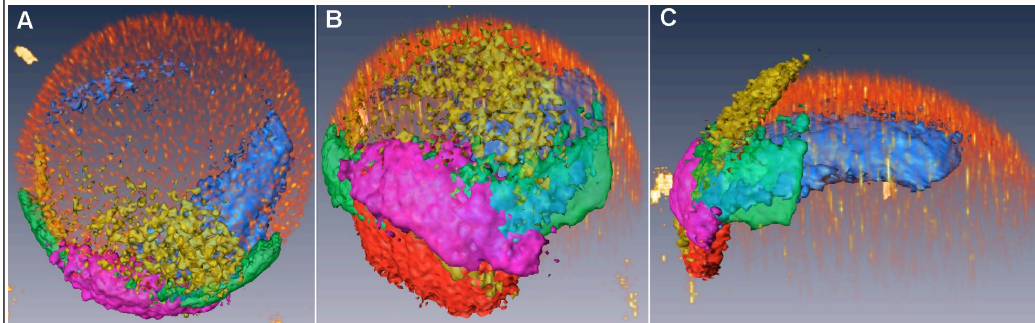






S. Desnoulez

Building gene expression patterns 4D atlas

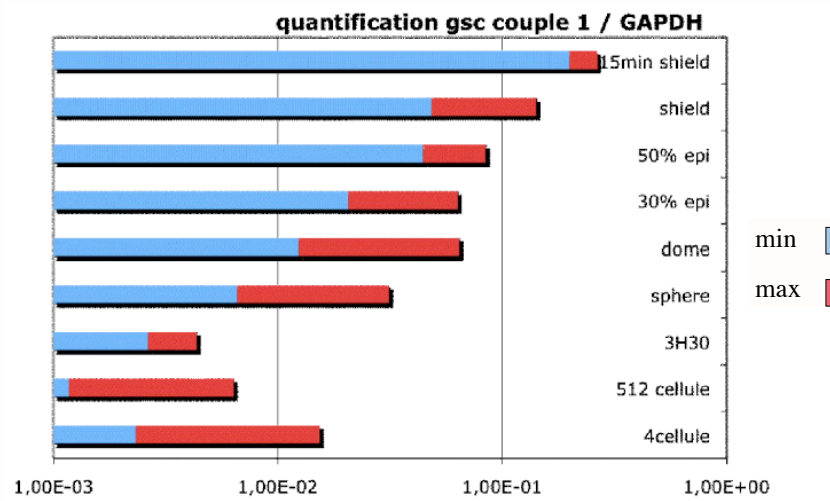


C. Castro, M. A. Luengo-Oroz, S. Desnoulez, L. Duloquin, L. Fernández-de-Manuel, M. J. Ledesma-Carbayo,
P. Bourguine, N. Peyrieras, A. Santos

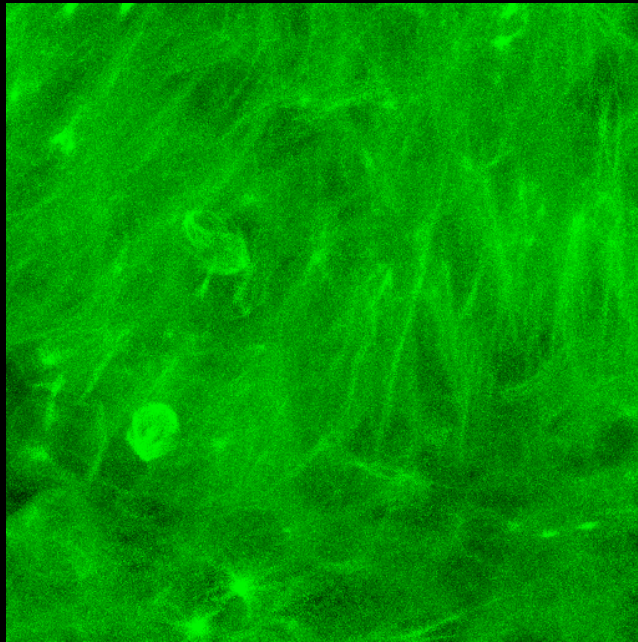
C. Castro, M. A. Luengo-Oroz, S. Desnoulez, L. Duloquin, L. Fernández-de-Manuel, M. J. Ledesma-Carbayo, P. Bourguine, N. Peyrieras, A. Santos and S. Montagna

Quantitative data to define models parameters

(Q RTPCR)



Sophie Desnoullez, coll. Georges Lutfalla



tubulin/eGFP fusion zebrafish larval brain

CNRS-CREA Paul Bourguine

INSERM Montpellier Georges Lutfalla

PHLAM Lille Pierre Suret

University of Bratislava Karol Mikula

UPM Madrid Andres Santos

LOB Polytechnique Emmanuel Beaurepaire

DENALI SA (Belgique) Dominique Snyers

CNRS-IDG Guy Wormser

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Institut Pasteur Jean-François Nicolas

University of Bologna Alessandro Sarti

EMBL Ernst Stelzer

Partners

